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Applicant(s): **Mariagrazia PIZZA, et al.**

Int'l Serial No.: PCT/US00/05928

Int'l Filing Date: 08 March 2000

Docket No.: **CHIR-0319**

Date Sent: October 30, 2001 DYT/MJR/vchicchi

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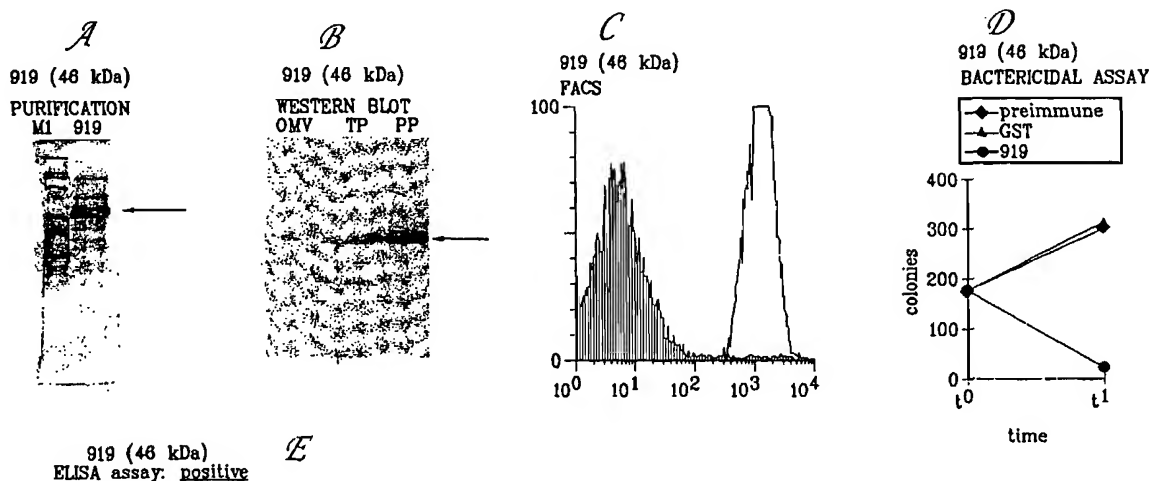
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<p>(21) International Application Number: PCT/US00/05928</p> <p>(22) International Filing Date: 8 March 2000 (08.03.00)</p> <p>(30) Priority Data:</p> <table border="0"> <tr> <td>60/132,068</td> <td>30 April 1999 (30.04.99)</td> <td>US</td> </tr> <tr> <td>PCT/US99/23573</td> <td>8 October 1999 (08.10.99)</td> <td>US</td> </tr> <tr> <td>0004695.3</td> <td>28 February 2000 (28.02.00)</td> <td>GB</td> </tr> </table> <p>(71) Applicants (for all designated States except US): CHIRON CORPORATION [US/US]; 4560 Horton Street, Emeryville, CA 94608 (US). THE INSTITUTE FOR GENOMIC RESEARCH [US/US]; 9212 Medical Center Drive, Rockville, MD 20850 (US).</p> <p>(72) Inventors; and</p> <p>(75) Inventors/Applicants (for US only): PIZZA, Mariagrazia [IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT). HICKEY, Erin [US/US]; 4569 Horton Street, Emeryville, CA 94608-2916 (US). PETERSON, Jeremy [US/US]; 4569 Horton Street, Emeryville, CA 94608-2916 (US). TETTELIN, Herve [US/US]; 4569 Horton Street, Emeryville, CA 94608-2916 (US). VENTER, J., Craig [US/US]; 4569 Horton Street, Emeryville, CA 94608-2916 (US). MASIGNANI, Vega [IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT). GALEOTTI, Cesira</p>	60/132,068	30 April 1999 (30.04.99)	US	PCT/US99/23573	8 October 1999 (08.10.99)	US	0004695.3	28 February 2000 (28.02.00)	GB	<p>[IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT). MORA, Marirosa [IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT). RATTI, Giulio [IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT). SCARSELLI, Maria [IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT). SCARLATO, Vincenzo [IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT). RAPPUOLI, Rino [IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT). FRAZER, Claire, M. [US/US]; 4560 Horton Street, Emeryville, CA 94608 (US). GRANDI, Guido [IT/IT]; Chiron SpA, Via Fiorentina 1, I-53100 Siena (IT).</p> <p>(74) Agent: HARBIN, Alisa, A.; Chiron Corporation, Intellectual Property - R440, P.O. Box 8097, Emeryville, CA 94662-8097 (US).</p> <p>(81) Designated States: AE, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, US, UZ, VN, YU, ZA, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SL, SZ, TZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).</p> <p>Published With international search report. Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</p>
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(54) Title: NEISSERIA GENOMIC SEQUENCES AND METHODS OF THEIR USE



(57) Abstract

The invention provides methods of obtaining immunogenic proteins from genomic sequences including *Neisseria*, including the amino acid sequences and the corresponding nucleotide sequences, as well as the genomic sequence of *Neisseria meningitidis B*. The proteins so obtained are useful antigens for vaccines, immunogenic compositions, and/or diagnostics.

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NEISSERIA GENOMIC SEQUENCES AND METHODS OF THEIR USE

This application claims priority to provisional U.S. application serial no. 60/132,068, filed 30 April 1999; PCT/US99/23573, filed 8 October 1999 (to be published April 2000); and Great Britain application serial no. GB-0004695.3, filed 28 February 2000.

This invention relates to methods of obtaining antigens and immunogens, the antigens and immunogens so obtained, and nucleic acids from the bacterial species: *Neisseria meningitidis*. In particular, it relates to genomic sequences from the bacterium; more particularly its "B" serogroup.

BACKGROUND

Neisseria meningitidis is a non-motile, gram negative diplococcus human pathogen. It colonizes the pharynx, causing meningitis and, occasionally, septicaemia in the absence of meningitis. It is closely related to *N. gonorrhoea*, although one feature that clearly differentiates meningococcus from gonococcus is the presence of a polysaccharide capsule that is present in all pathogenic meningococci.

N. meningitidis causes both endemic and epidemic disease. In the United States the attack rate is 0.6-1 per 100,000 persons per year, and it can be much greater during outbreaks. (see Lieberman *et al.* (1996) Safety and Immunogenicity of a Serogroups A/C *Neisseria meningitidis* Oligosaccharide-Protein Conjugate Vaccine in Young Children. *JAMA* 275(19):1499-1503; Schuchat *et al* (1997) Bacterial Meningitis in the United States in 1995. *N Engl J Med* 337(14):970-976). In developing countries, endemic disease rates are much higher and during epidemics incidence rates can reach 500 cases per 100,000 persons per year. Mortality is extremely high, at 10-20% in the United States, and much higher in developing countries. Following the introduction of the conjugate vaccine against *Haemophilus influenzae*, *N. meningitidis* is the major cause of bacterial meningitis at all ages in the United States (Schuchat *et al* (1997) *supra*).

Based on the organism's capsular polysaccharide, 12 serogroups of *N. meningitidis* have been identified. Group A is the pathogen most often implicated in epidemic disease in sub-Saharan Africa. Serogroups B and C are responsible for the vast majority of cases in the

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United States and in most developed countries. Serogroups W135 and Y are responsible for the rest of the cases in the United States and developed countries. The meningococcal vaccine currently in use is a tetravalent polysaccharide vaccine composed of serogroups A, C, Y and W135. Although efficacious in adolescents and adults, it induces a poor immune response and short duration of protection, and cannot be used in infants (e.g., Morbidity and Mortality weekly report, Vol. 46, No. RR-5 (1997)). This is because polysaccharides are T-cell independent antigens that induce a weak immune response that cannot be boosted by repeated immunization. Following the success of the vaccination against *H. influenzae*, conjugate vaccines against serogroups A and C have been developed and are at the final stage of clinical testing (Zollinger WD "New and Improved Vaccines Against Meningococcal Disease". In: *New Generation Vaccines*, supra, pp. 469-488; Lieberman *et al* (1996) supra; Costantino *et al* (1992) Development and phase I clinical testing of a conjugate vaccine against meningococcus A (menA) and C (menC) (*Vaccine* 10:691-698)).

Meningococcus B (MenB) remains a problem, however. This serotype currently is responsible for approximately 50% of total meningitis in the United States, Europe, and South America. The polysaccharide approach cannot be used because the MenB capsular polysaccharide is a polymer of $\alpha(2-8)$ -linked *N*-acetyl neuraminic acid that is also present in mammalian tissue. This results in tolerance to the antigen; indeed, if an immune response were elicited, it would be anti-self, and therefore undesirable. In order to avoid induction of autoimmunity and to induce a protective immune response, the capsular polysaccharide has, for instance, been chemically modified substituting the *N*-acetyl groups with *N*-propionyl groups, leaving the specific antigenicity unaltered (Romero & Outschoorn (1994) Current status of Meningococcal group B vaccine candidates: capsular or non-capsular? *Clin Microbiol Rev* 7(4):559-575).

Alternative approaches to MenB vaccines have used complex mixtures of outer membrane proteins (OMPs), containing either the OMPs alone, or OMPs enriched in porins, or deleted of the class 4 OMPs that are believed to induce antibodies that block bactericidal activity. This approach produces vaccines that are not well characterized. They are able to protect against the homologous strain, but are not effective at large where there are many antigenic variants of the outer membrane proteins. To overcome the antigenic variability, multivalent vaccines containing up to nine different porins have been constructed (e.g.,

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Poolman JT (1992) Development of a meningococcal vaccine. *Infect. Agents Dis.* 4:13-28). Additional proteins to be used in outer membrane vaccines have been the opa and opc proteins, but none of these approaches have been able to overcome the antigenic variability (e.g., Ala'Aldeen & Borriello (1996) The meningococcal transferrin-binding proteins 1 and 2 are both surface exposed and generate bactericidal antibodies capable of killing homologous and heterologous strains. *Vaccine* 14(1):49-53).

A certain amount of sequence data is available for meningococcal and gonococcal genes and proteins (e.g., EP-A-0467714, WO96/29412), but this is by no means complete. The provision of further sequences could provide an opportunity to identify secreted or surface-exposed proteins that are presumed targets for the immune system and which are not antigenically variable or at least are more antigenically conserved than other and more variable regions. Thus, those antigenic sequences that are more highly conserved are preferred sequences. Those sequences specific to *Neisseria meningitidis* or *Neisseria gonorrhoeae* that are more highly conserved are further preferred sequences. For instance, some of the identified proteins could be components of efficacious vaccines against meningococcus B, some could be components of vaccines against all meningococcal serotypes, and others could be components of vaccines against all pathogenic *Neisseriae*. The identification of sequences from the bacterium will also facilitate the production of biological probes, particularly organism-specific probes.

It is thus an object of the invention is to provide Neisserial DNA sequences which (1) encode proteins predicted and/or shown to be antigenic or immunogenic, (2) can be used as probes or amplification primers, and (3) can be analyzed by bioinformatics.

BRIEF DESCRIPTION OF THE DRAWINGS

Fig. 1 illustrates the products of protein expression and purification of the predicted ORF 919 as cloned and expressed in *E. coli*.

Fig. 2 illustrates the products of protein expression and purification of the predicted ORF 279 as cloned and expressed in *E. coli*.

Fig. 3 illustrates the products of protein expression and purification of the predicted ORF 576-1 as cloned and expressed in *E. coli*.

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Fig. 4 illustrates the products of protein expression and purification of the predicted ORF 519-1 as cloned and expressed in *E. coli*.

Fig. 5 illustrates the products of protein expression and purification of the predicted ORF 121-1 as cloned and expressed in *E. coli*.

Fig. 6 illustrates the products of protein expression and purification of the predicted ORF 128-1 as cloned and expressed in *E. coli*.

Fig. 7 illustrates the products of protein expression and purification of the predicted ORF 206 as cloned and expressed in *E. coli*.

Fig. 8 illustrates the products of protein expression and purification of the predicted ORF 287 as cloned and expressed in *E. coli*.

Fig. 9 illustrates the products of protein expression and purification of the predicted ORF 406 as cloned and expressed in *E. coli*.

Fig. 10 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 919 as cloned and expressed in *E. coli*.

Fig. 11 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 279 as cloned and expressed in *E. coli*.

Fig. 12 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 576-1 as cloned and expressed in *E. coli*.

Fig. 13 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 519-1 as cloned and expressed in *E. coli*.

Fig. 14 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 121-1 as cloned and expressed in *E. coli*.

Fig. 15 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 128-1 as cloned and expressed in *E. coli*.

Fig. 16 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 206 as cloned and expressed in *E. coli*.

Fig. 17 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 287 as cloned and expressed in *E. coli*.

Fig. 18 illustrates the hydrophilicity plot, antigenic index and AMPHI regions of the products of protein expression the predicted ORF 406 as cloned and expressed in *E. coli*.

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THE INVENTION

The first complete sequence of the genome of *N. meningitidis* was disclosed as 961 partial contiguous nucleotide sequences, shown as SEQ ID NOs:1-961 of co-owned PCT/US99/23573 (the '573 application), filed 8 October 1999 (to be published April 2000). A single sequence full length genome of *N. meningitidis* was also disclosed as SEQ ID NO. 1068 of the '573 application. The invention is based on a full length genome of *N. meningitidis* which appears as SEQ ID NO. 1 in the present application as Appendix A hereto. The 961 sequences of the '573 application represent substantially the whole genome of serotype B of *N. meningitidis* (>99.98%). There is partial overlap between some of the 961 contiguous sequences ("contigs") shown in the 961 sequences, which overlap was used to construct the single full length sequence shown in SEQ ID NO. 1 in Appendix A hereto, using the TIGR Assembler [G.S. Sutton et al., *TIGR Assembler: A New Tool for Assembling Large Shotgun Sequencing Projects*, Genome Science and Technology, 1:9-19 (1995)]. Some of the nucleotides in the contigs had been previously released. (See ftp://ftp.tigr.org/pub/data/n_meningitidis on the world-wide web or "WWW"). The coordinates of the 2508 released sequences in the present contigs are presented in Appendix A of the '573 application. These data include the contig number (or i.d.) as presented in the first column; the name of the sequence as found on WWW is in the second column; with the coordinates of the contigs in the third and fourth columns, respectively. The sequences of certain MenB ORFs presented in Appendix B of the '573 application feature in International Patent Application filed by Chiron SpA on October 9, 1998 (PCT/IB98/01665) and January 14, 1999 (PCT/IB99/00103) respectively. Appendix B hereto provides a listing of 2158 open reading frames contained within the full length sequence found in SEQ ID NO. 1 in Appendix A hereto. The information set forth in Appendix B hereto includes the "NMB" name of the sequence, the putative translation product, and the beginning and ending nucleotide positions within SEQ ID NO. 1 which comprise the open reading frames. These open reading frames are referred to herein as the "NMB open reading frames".

In a first aspect, the invention provides nucleic acid including the *N. meningitidis* nucleotide sequence shown in SEQ ID NO. 1 in Appendix A hereto. It also provides nucleic acid comprising sequences having sequence identity to the nucleotide sequence disclosed herein. Depending on the particular sequence, the degree of sequence identity is preferably

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greater than 50% (e.g., 60%, 70%, 80%, 90%, 95%, 99% or more). These sequences include, for instance, mutants and allelic variants. The degree of sequence identity cited herein is determined across the length of the sequence determined by the Smith-Waterman homology search algorithm as implemented in MPSRCH program (Oxford Molecular) using an affine gap search with the following parameters: gap open penalty 12, gap extension penalty 1.

The invention also provides nucleic acid including a fragment of one or more of the nucleotide sequences set out herein, including the NMB open reading frames shown in Appendix B hereto. The fragment should comprise at least n consecutive nucleotides from the sequences and, depending on the particular sequence, n is 10 or more (e.g., 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 30, 35, 40, 45, 50, 60, 75, 100 or more). Preferably, the fragment is unique to the genome of *N. meningitidis*, that is to say it is not present in the genome of another organism. More preferably, the fragment is unique to the genome of strain B of *N. meningitidis*. The invention also provides nucleic acid that hybridizes to those provided herein. Conditions for hybridizing are disclosed herein.

The invention also provides nucleic acid including sequences complementary to those described above (e.g., for antisense, for probes, or for amplification primers).

Nucleic acid according to the invention can, of course, be prepared in many ways (e.g., by chemical synthesis, from DNA libraries, from the organism itself, etc.) and can take various forms (e.g., single-stranded, double-stranded, vectors, probes, primers, etc.). The term "nucleic acid" includes DNA and RNA, and also their analogs, such as those containing modified backbones, and also peptide nucleic acid (PNA) etc.

It will be appreciated that, as SEQ ID NOs:1-961 of the '573 application represent the substantially complete genome of the organism, with partial overlap, references to SEQ ID NOs:1-961 of the '573 application include within their scope references to the complete genomic sequence, that is, SEQ ID NO. 1 hereof. For example, where two SEQ ID NOs overlap, the invention encompasses the single sequence which is formed by assembling the two overlapping sequences, which full sequence will be found in SEQ ID NO. 1 hereof. Thus, for instance, a nucleotide sequence which bridges two SEQ ID NOs but is not present in its entirety in either SEQ ID NO is still within the scope of the invention. Such a sequence will be present in its entirety in the single full length sequence of SEQ ID NO. 1 of the present application.

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The invention also provides vectors including nucleotide sequences of the invention (e.g., expression vectors, sequencing vectors, cloning vectors, etc.) and host cells transformed with such vectors.

According to a further aspect, the invention provides a protein including an amino acid sequence encoded within a *N. meningitidis* nucleotide sequence set out herein. It also provides proteins comprising sequences having sequence identity to those proteins. Depending on the particular sequence, the degree of sequence identity is preferably greater than 50% (e.g., 60%, 70%, 80%, 90%, 95%, 99% or more). Sequence identity is determined as above disclosed. These homologous proteins include mutants and allelic variants, encoded within the *N. meningitidis* nucleotide sequence set out herein.

The invention further provides proteins including fragments of an amino acid sequence encoded within a *N. meningitidis* nucleotide sequence set out in the sequence listing. The fragments should comprise at least *n* consecutive amino acids from the sequences and, depending on the particular sequence, *n* is 7 or more (e.g., 8, 10, 12, 14, 16, 18, 20 or more). Preferably the fragments comprise an epitope from the sequence.

The proteins of the invention can, of course, be prepared by various means (e.g., recombinant expression, purification from cell culture, chemical synthesis, etc.) and in various forms (e.g. native, fusions etc.). They are preferably prepared in substantially isolated form (i.e., substantially free from other *N. meningitidis* host cell proteins).

Various tests can be used to assess the *in vivo* immunogenicity of the proteins of the invention. For example, the proteins can be expressed recombinantly or chemically synthesized and used to screen patient sera by immunoblot. A positive reaction between the protein and patient serum indicates that the patient has previously mounted an immune response to the protein in question; i.e., the protein is an immunogen. This method can also be used to identify immunodominant proteins.

The invention also provides nucleic acid encoding a protein of the invention.

In a further aspect, the invention provides a computer, a computer memory, a computer storage medium (e.g., floppy disk, fixed disk, CD-ROM, etc.), and/or a computer database containing the nucleotide sequence of nucleic acid according to the invention. Preferably, it contains one or more of the *N. meningitidis* nucleotide sequences set out herein.

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This may be used in the analysis of the *N. meningitidis* nucleotide sequences set out herein. For instance, it may be used in a search to identify open reading frames (ORFs) or coding sequences within the sequences.

In a further aspect, the invention provides a method for identifying an amino acid sequence, comprising the step of searching for putative open reading frames or protein-coding sequences within a *N. meningitidis* nucleotide sequence set out herein. Similarly, the invention provides the use of a *N. meningitidis* nucleotide sequence set out herein in a search for putative open reading frames or protein-coding sequences.

Open-reading frame or protein-coding sequence analysis is generally performed on a computer using standard bioinformatic techniques. Typical algorithms or program used in the analysis include ORFFINDER (NCBI), GENMARK [Borodovsky & McIninch (1993) *Computers Chem* 17:122-133], and GLIMMER [Salzberg et al. (1998) *Nucl Acids Res* 26:544-548].

A search for an open reading frame or protein-coding sequence may comprise the steps of searching a *N. meningitidis* nucleotide sequence set out herein for an initiation codon and searching the upstream sequence for an in-frame termination codon. The intervening codons represent a putative protein-coding sequence. Typically, all six possible reading frames of a sequence will be searched.

An amino acid sequence identified in this way can be expressed using any suitable system to give a protein. This protein can be used to raise antibodies which recognize epitopes within the identified amino acid sequence. These antibodies can be used to screen *N. meningitidis* to detect the presence of a protein comprising the identified amino acid sequence.

Furthermore, once an ORF or protein-coding sequence is identified, the sequence can be compared with sequence databases. Sequence analysis tools can be found at NCBI (<http://www.ncbi.nlm.nih.gov>) e.g., the algorithms BLAST, BLAST2, BLASTn, BLASTp, tBLASTn, BLASTx, & tBLASTx [see also Altschul *et al.* (1997) Gapped BLAST and PSI-BLAST: new generation of protein database search programs. *Nucleic Acids Research* 25:2289-3402]. Suitable databases for comparison include the nonredundant GenBank, EMBL, DDBJ and PDB sequences, and the nonredundant GenBank CDS translations, PDB,

SwissProt, Spupdate and PIR sequences. This comparison may give an indication of the function of a protein.

Hydrophobic domains in an amino acid sequence can be predicted using algorithms such as those based on the statistical studies of Esposti *et al.* [Critical evaluation of the hydropathy of membrane proteins (1990) *Eur J Biochem* 190:207-219]. Hydrophobic domains represent potential transmembrane regions or hydrophobic leader sequences, which suggest that the proteins may be secreted or be surface-located. These properties are typically representative of good immunogens.

Similarly, transmembrane domains or leader sequences can be predicted using the PSORT algorithm (<http://www.psort.nibb.ac.jp>), and functional domains can be predicted using the MOTIFS program (GCG Wisconsin & PROSITE).

The invention also provides nucleic acid including an open reading frame or protein-coding sequence present in a *N. meningitidis* nucleotide sequence set out herein. Furthermore, the invention provides a protein including the amino acid sequence encoded by this open reading frame or protein-coding sequence.

According to a further aspect, the invention provides antibodies which bind to these proteins. These may be polyclonal or monoclonal and may be produced by any suitable means known to those skilled in the art.

The antibodies of the invention can be used in a variety of ways, e.g., for confirmation that a protein is expressed, or to confirm where a protein is expressed. Labeled antibody (e.g., fluorescent labeling for FACS) can be incubated with intact bacteria and the presence of label on the bacterial surface confirms the location of the protein, for instance.

According to a further aspect, the invention provides compositions including protein, antibody, and/or nucleic acid according to the invention. These compositions may be suitable as vaccines, as immunogenic compositions, or as diagnostic reagents.

The invention also provides nucleic acid, protein, or antibody according to the invention for use as medicaments (e.g., as vaccines) or as diagnostic reagents. It also provides the use of nucleic acid, protein, or antibody according to the invention in the manufacture of (i) a medicament for treating or preventing infection due to Neisserial bacteria (ii) a diagnostic reagent for detecting the presence of Neisserial bacteria or of antibodies raised against Neisserial bacteria. Said Neisserial bacteria may be any species or

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strain (such as *N. gonorrhoeae*) but are preferably *N. meningitidis*, especially strain A, strain B or strain C.

In still yet another aspect, the present invention provides for compositions including proteins, nucleic acid molecules, or antibodies. More preferable aspects of the present invention are drawn to immunogenic compositions of proteins. Further preferable aspects of the present invention contemplate pharmaceutical immunogenic compositions of proteins or vaccines and the use thereof in the manufacture of a medicament for the treatment or prevention of infection due to Neisserial bacteria, preferably infection of MenB.

The invention also provides a method of treating a patient, comprising administering to the patient a therapeutically effective amount of nucleic acid, protein, and/or antibody according to the invention.

According to further aspects, the invention provides various processes.

A process for producing proteins of the invention is provided, comprising the step of culturing a host cell according to the invention under conditions which induce protein expression. A process which may further include chemical synthesis of proteins and/or chemical synthesis (at least in part) of nucleotides.

A process for detecting polynucleotides of the invention is provided, comprising the steps of: (a) contacting a nucleic probe according to the invention with a biological sample under hybridizing conditions to form duplexes; and (b) detecting said duplexes.

A process for detecting proteins of the invention is provided, comprising the steps of: (a) contacting an antibody according to the invention with a biological sample under conditions suitable for the formation of an antibody-antigen complexes; and (b) detecting said complexes.

Another aspect of the present invention provides for a process for detecting antibodies that selectably bind to antigens or polypeptides or proteins specific to any species or strain of Neisserial bacteria and preferably to strains of *N. gonorrhoeae* but more preferably to strains of *N. meningitidis*, especially strain A, strain B or strain C, more preferably MenB, where the process comprises the steps of: (a) contacting antigen or polypeptide or protein according to the invention with a biological sample under conditions suitable for the formation of an antibody-antigen complexes; and (b) detecting said complexes.

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Having now generally described the invention, the same will be more readily understood through reference to the following examples which are provided by way of illustration, and are not intended to be limiting of the present invention, unless specified.

Methodology - Summary of standard procedures and techniques.

General

This invention provides *Neisseria meningitidis* MenB nucleotide sequences, amino acid sequences encoded therein. With these disclosed sequences, nucleic acid probe assays and expression cassettes and vectors can be produced. The proteins can also be chemically synthesized. The expression vectors can be transformed into host cells to produce proteins. The purified or isolated polypeptides can be used to produce antibodies to detect MenB proteins. Also, the host cells or extracts can be utilized for biological assays to isolate agonists or antagonists. In addition, with these sequences one can search to identify open reading frames and identify amino acid sequences. The proteins may also be used in immunogenic compositions and as vaccine components.

The practice of the present invention will employ, unless otherwise indicated, conventional techniques of molecular biology, microbiology, recombinant DNA, and immunology, which are within the skill of the art. Such techniques are explained fully in the literature e.g., Sambrook *Molecular Cloning: A Laboratory Manual, Second Edition* (1989); *DNA Cloning, Volumes I and ii* (D.N Glover ed. 1985); *Oligonucleotide Synthesis* (M.J. Gait ed, 1984); *Nucleic Acid Hybridization* (B.D. Hames & S.J. Higgins eds. 1984); *Transcription and Translation* (B.D. Hames & S.J. Higgins eds. 1984); *Animal Cell Culture* (R.I. Freshney ed. 1986); *Immobilized Cells and Enzymes* (IRL Press, 1986); B. Perbal, *A Practical Guide to Molecular Cloning* (1984); the *Methods in Enzymology* series (Academic Press, Inc.), especially volumes 154 & 155; *Gene Transfer Vectors for Mammalian Cells* (J.H. Miller and M.P. Calos eds. 1987, Cold Spring Harbor Laboratory); Mayer and Walker, eds. (1987), *Immunochemical Methods in Cell and Molecular Biology* (Academic Press, London); Scopes, (1987) *Protein Purification: Principles and Practice*, Second Edition (Springer-Verlag, N.Y.), and *Handbook of Experimental Immunology, Volumes I-IV* (D.M. Weir and C.C. Blackwell eds 1986).

Standard abbreviations for nucleotides and amino acids are used in this specification.

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All publications, patents, and patent applications cited herein are incorporated in full by reference.

Expression systems

The *Neisseria* MenB nucleotide sequences can be expressed in a variety of different expression systems; for example those used with mammalian cells, plant cells, baculoviruses, bacteria, and yeast.

i. Mammalian Systems

Mammalian expression systems are known in the art. A mammalian promoter is any DNA sequence capable of binding mammalian RNA polymerase and initiating the downstream (3') transcription of a coding sequence (e.g., structural gene) into mRNA. A promoter will have a transcription initiating region, which is usually placed proximal to the 5' end of the coding sequence, and a TATA box, usually located 25-30 base pairs (bp) upstream of the transcription initiation site. The TATA box is thought to direct RNA polymerase II to begin RNA synthesis at the correct site. A mammalian promoter will also contain an upstream promoter element, usually located within 100 to 200 bp upstream of the TATA box. An upstream promoter element determines the rate at which transcription is initiated and can act in either orientation (Sambrook et al. (1989) "Expression of Cloned Genes in Mammalian Cells." In *Molecular Cloning: A Laboratory Manual*, 2nd ed.).

Mammalian viral genes are often highly expressed and have a broad host range; therefore sequences encoding mammalian viral genes provide particularly useful promoter sequences. Examples include the SV40 early promoter, mouse mammary tumor virus LTR promoter, adenovirus major late promoter (Ad MLP), and herpes simplex virus promoter. In addition, sequences derived from non-viral genes, such as the murine metallothionein gene, also provide useful promoter sequences. Expression may be either constitutive or regulated (inducible). Depending on the promoter selected, many promoters may be inducible using known substrates, such as the use of the mouse mammary tumor virus (MMTV) promoter with the glucocorticoid responsive element (GRE) that is induced by glucocorticoid in hormone-responsive transformed cells (see for example, U.S. Patent 5,783,681).

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The presence of an enhancer element (enhancer), combined with the promoter elements described above, will usually increase expression levels. An enhancer is a regulatory DNA sequence that can stimulate transcription up to 1000-fold when linked to homologous or heterologous promoters, with synthesis beginning at the normal RNA start site. Enhancers are also active when they are placed upstream or downstream from the transcription initiation site, in either normal or flipped orientation, or at a distance of more than 1000 nucleotides from the promoter (Maniatis et al. (1987) *Science* 236:1237; Alberts et al. (1989) *Molecular Biology of the Cell*, 2nd ed.). Enhancer elements derived from viruses may be particularly useful, because they usually have a broader host range. Examples include the SV40 early gene enhancer (Dijkema et al (1985) *EMBO J.* 4:761) and the enhancer/promoters derived from the long terminal repeat (LTR) of the Rous Sarcoma Virus (Gorman et al. (1982b) *Proc. Natl. Acad. Sci.* 79:6777) and from human cytomegalovirus (Boshart et al. (1985) *Cell* 41:521). Additionally, some enhancers are regulatable and become active only in the presence of an inducer, such as a hormone or metal ion (Sassone-Corsi and Borelli (1986) *Trends Genet.* 2:215; Maniatis et al. (1987) *Science* 236:1237).

A DNA molecule may be expressed intracellularly in mammalian cells. A promoter sequence may be directly linked with the DNA molecule, in which case the first amino acid at the N-terminus of the recombinant protein will always be a methionine, which is encoded by the ATG start codon. If desired, the N-terminus may be cleaved from the protein by *in vitro* incubation with cyanogen bromide.

Alternatively, foreign proteins can also be secreted from the cell into the growth media by creating chimeric DNA molecules that encode a fusion protein comprised of a leader sequence fragment that provides for secretion of the foreign protein in mammalian cells. Preferably, there are processing sites encoded between the leader fragment and the foreign gene that can be cleaved either *in vivo* or *in vitro*. The leader sequence fragment usually encodes a signal peptide comprised of hydrophobic amino acids which direct the secretion of the protein from the cell. The adenovirus tripartite leader is an example of a leader sequence that provides for secretion of a foreign protein in mammalian cells.

Usually, transcription termination and polyadenylation sequences recognized by mammalian cells are regulatory regions located 3' to the translation stop codon and thus, together with the promoter elements, flank the coding sequence. The 3' terminus of the

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mature mRNA is formed by site-specific post-transcriptional cleavage and polyadenylation (Birnstiel et al. (1985) *Cell* 41:349; Proudfoot and Whitelaw (1988) "Termination and 3' end processing of eukaryotic RNA. In *Transcription and splicing* (ed. B.D. Hames and D.M. Glover); Proudfoot (1989) *Trends Biochem. Sci.* 14:105). These sequences direct the transcription of an mRNA which can be translated into the polypeptide encoded by the DNA. Examples of transcription terminator/polyadenylation signals include those derived from SV40 (Sambrook et al (1989) "Expression of cloned genes in cultured mammalian cells." In *Molecular Cloning: A Laboratory Manual*).

Usually, the above-described components, comprising a promoter, polyadenylation signal, and transcription termination sequence are put together into expression constructs. Enhancers, introns with functional splice donor and acceptor sites, and leader sequences may also be included in an expression construct, if desired. Expression constructs are often maintained in a replicon, such as an extrachromosomal element (e.g., plasmids) capable of stable maintenance in a host, such as mammalian cells or bacteria. Mammalian replication systems include those derived from animal viruses, which require trans-acting factors to replicate. For example, plasmids containing the replication systems of papovaviruses, such as SV40 (Gluzman (1981) *Cell* 23:175) or polyomavirus, replicate to extremely high copy number in the presence of the appropriate viral T antigen. Additional examples of mammalian replicons include those derived from bovine papillomavirus and Epstein-Barr virus. Additionally, the replicon may have two replication systems, thus allowing it to be maintained, for example, in mammalian cells for expression and in a prokaryotic host for cloning and amplification. Examples of such mammalian-bacteria shuttle vectors include pMT2 (Kaufman et al. (1989) *Mol. Cell. Biol.* 9:946) and pHEBO (Shimizu et al. (1986) *Mol. Cell. Biol.* 6:1074).

The transformation procedure used depends upon the host to be transformed. Methods for introduction of heterologous polynucleotides into mammalian cells are known in the art and include dextran-mediated transfection, calcium phosphate precipitation, polybrene mediated transfection, protoplast fusion, electroporation, encapsulation of the polynucleotide(s) in liposomes, and direct microinjection of the DNA into nuclei.

Mammalian cell lines available as hosts for expression are known in the art and include many immortalized cell lines available from the American Type Culture Collection

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(ATCC), including but not limited to, Chinese hamster ovary (CHO) cells, HeLa cells, baby hamster kidney (BHK) cells, monkey kidney cells (COS), human hepatocellular carcinoma cells (e.g., Hep G2), and a number of other cell lines.

ii. Plant Cellular Expression Systems

There are many plant cell culture and whole plant genetic expression systems known in the art. Exemplary plant cellular genetic expression systems include those described in patents, such as: U.S. 5,693,506; US 5,659,122; and US 5,608,143. Additional examples of genetic expression in plant cell culture has been described by Zenk, *Phytochemistry* 30:3861-3863 (1991). Descriptions of plant protein signal peptides may be found in addition to the references described above in Vaulcombe et al., *Mol. Gen. Genet.* 209:33-40 (1987); Chandler et al., *Plant Molecular Biology* 3:407-418 (1984); Rogers, *J. Biol. Chem.* 260:3731-3738 (1985); Rothstein et al., *Gene* 55:353-356 (1987); Whittier et al., *Nucleic Acids Research* 15:2515-2535 (1987); Wirsal et al., *Molecular Microbiology* 3:3-14 (1989); Yu et al., *Gene* 122:247-253 (1992). A description of the regulation of plant gene expression by the phytohormone, gibberellic acid and secreted enzymes induced by gibberellic acid can be found in R.L. Jones and J. MacMillin, Gibberellins: in: *Advanced Plant Physiology*,. Malcolm B. Wilkins, ed., 1984 Pitman Publishing Limited, London, pp. 21-52. References that describe other metabolically-regulated genes: Sheen, *Plant Cell*, 2:1027-1038(1990); Maas et al., *EMBO J.* 9:3447-3452 (1990); Benkel and Hickey, *Proc. Natl. Acad. Sci.* 84:1337-1339 (1987)

Typically, using techniques known in the art, a desired polynucleotide sequence is inserted into an expression cassette comprising genetic regulatory elements designed for operation in plants. The expression cassette is inserted into a desired expression vector with companion sequences upstream and downstream from the expression cassette suitable for expression in a plant host. The companion sequences will be of plasmid or viral origin and provide necessary characteristics to the vector to permit the vectors to move DNA from an original cloning host, such as bacteria, to the desired plant host. The basic bacterial/plant vector construct will preferably provide a broad host range prokaryote replication origin; a prokaryote selectable marker; and, for *Agrobacterium* transformations, T DNA sequences for *Agrobacterium*-mediated transfer to plant chromosomes. Where the heterologous gene is not

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readily amenable to detection, the construct will preferably also have a selectable marker gene suitable for determining if a plant cell has been transformed. A general review of suitable markers, for example for the members of the grass family, is found in Wilmink and Dons, 1993, *Plant Mol. Biol. Repr*, 11(2):165-185.

Sequences suitable for permitting integration of the heterologous sequence into the plant genome are also recommended. These might include transposon sequences and the like for homologous recombination as well as Ti sequences which permit random insertion of a heterologous expression cassette into a plant genome. Suitable prokaryote selectable markers include resistance toward antibiotics such as ampicillin or tetracycline. Other DNA sequences encoding additional functions may also be present in the vector, as is known in the art.

The nucleic acid molecules of the subject invention may be included into an expression cassette for expression of the protein(s) of interest. Usually, there will be only one expression cassette, although two or more are feasible. The recombinant expression cassette will contain in addition to the heterologous protein encoding sequence the following elements, a promoter region, plant 5' untranslated sequences, initiation codon depending upon whether or not the structural gene comes equipped with one, and a transcription and translation termination sequence. Unique restriction enzyme sites at the 5' and 3' ends of the cassette allow for easy insertion into a pre-existing vector.

A heterologous coding sequence may be for any protein relating to the present invention. The sequence encoding the protein of interest will encode a signal peptide which allows processing and translocation of the protein, as appropriate, and will usually lack any sequence which might result in the binding of the desired protein of the invention to a membrane. Since, for the most part, the transcriptional initiation region will be for a gene which is expressed and translocated during germination, by employing the signal peptide which provides for translocation, one may also provide for translocation of the protein of interest. In this way, the protein(s) of interest will be translocated from the cells in which they are expressed and may be efficiently harvested. Typically secretion in seeds are across the aleurone or scutellar epithelium layer into the endosperm of the seed. While it is not required that the protein be secreted from the cells in which the protein is produced, this facilitates the isolation and purification of the recombinant protein.

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Since the ultimate expression of the desired gene product will be in a eucaryotic cell it is desirable to determine whether any portion of the cloned gene contains sequences which will be processed out as introns by the host's splicosome machinery. If so, site-directed mutagenesis of the "intron" region may be conducted to prevent losing a portion of the genetic message as a false intron code, Reed and Maniatis, *Cell* 41:95-105, 1985.

The vector can be microinjected directly into plant cells by use of micropipettes to mechanically transfer the recombinant DNA. Crossway, *Mol. Gen. Genet*, 202:179-185, 1985. The genetic material may also be transferred into the plant cell by using polyethylene glycol, Krens, et al., *Nature*, 296, 72-74, 1982. Another method of introduction of nucleic acid segments is high velocity ballistic penetration by small particles with the nucleic acid either within the matrix of small beads or particles, or on the surface, Klein, et al., *Nature*, 327, 70-73, 1987 and Knudsen and Muller, 1991, *Planta*, 185:330-336 teaching particle bombardment of barley endosperm to create transgenic barley. Yet another method of introduction would be fusion of protoplasts with other entities, either minicells, cells, lysosomes or other fusible lipid-surfaced bodies, Fraley, et al., *Proc. Natl. Acad. Sci. USA*, 79, 1859-1863, 1982.

The vector may also be introduced into the plant cells by electroporation. (Fromm et al., *Proc. Natl Acad. Sci. USA* 82:5824, 1985). In this technique, plant protoplasts are electroporated in the presence of plasmids containing the gene construct. Electrical impulses of high field strength reversibly permeabilize biomembranes allowing the introduction of the plasmids. Electroporated plant protoplasts reform the cell wall, divide, and form plant callus.

All plants from which protoplasts can be isolated and cultured to give whole regenerated plants can be transformed by the present invention so that whole plants are recovered which contain the transferred gene. It is known that practically all plants can be regenerated from cultured cells or tissues, including but not limited to all major species of sugarcane, sugar beet, cotton, fruit and other trees, legumes and vegetables. Some suitable plants include, for example, species from the genera *Fragaria*, *Lotus*, *Medicago*, *Onobrychis*, *Trifolium*, *Trigonella*, *Vigna*, *Citrus*, *Linum*, *Geranium*, *Manihot*, *Daucus*, *Arabidopsis*, *Brassica*, *Raphanus*, *Sinapis*, *Atropa*, *Capsicum*, *Datura*, *Hyoscyamus*, *Lycopersion*, *Nicotiana*, *Solanum*, *Petunia*, *Digitalis*, *Majorana*, *Cichorium*, *Helianthus*, *Lactuca*, *Bromus*, *Asparagus*, *Antirrhinum*, *Hererocallis*, *Nemesia*, *Pelargonium*, *Panicum*, *Pennisetum*,

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Ranunculus, Senecio, Salpiglossis, Cucumis, Browaalia, Glycine, Lolium, Zea, Triticum, Sorghum, and Datura.

Means for regeneration vary from species to species of plants, but generally a suspension of transformed protoplasts containing copies of the heterologous gene is first provided. Callus tissue is formed and shoots may be induced from callus and subsequently rooted. Alternatively, embryo formation can be induced from the protoplast suspension. These embryos germinate as natural embryos to form plants. The culture media will generally contain various amino acids and hormones, such as auxin and cytokinins. It is also advantageous to add glutamic acid and proline to the medium, especially for such species as corn and alfalfa. Shoots and roots normally develop simultaneously. Efficient regeneration will depend on the medium, on the genotype, and on the history of the culture. If these three variables are controlled, then regeneration is fully reproducible and repeatable.

In some plant cell culture systems, the desired protein of the invention may be excreted or alternatively, the protein may be extracted from the whole plant. Where the desired protein of the invention is secreted into the medium, it may be collected. Alternatively, the embryos and embryoless-half seeds or other plant tissue may be mechanically disrupted to release any secreted protein between cells and tissues. The mixture may be suspended in a buffer solution to retrieve soluble proteins. Conventional protein isolation and purification methods will be then used to purify the recombinant protein. Parameters of time, temperature pH, oxygen, and volumes will be adjusted through routine methods to optimize expression and recovery of heterologous protein.

iii. Baculovirus Systems

The polynucleotide encoding the protein can also be inserted into a suitable insect expression vector, and is operably linked to the control elements within that vector. Vector construction employs techniques which are known in the art. Generally, the components of the expression system include a transfer vector, usually a bacterial plasmid, which contains both a fragment of the baculovirus genome, and a convenient restriction site for insertion of the heterologous gene or genes to be expressed; a wild type baculovirus with a sequence homologous to the baculovirus-specific fragment in the transfer vector (this allows for the

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homologous recombination of the heterologous gene in to the baculovirus genome); and appropriate insect host cells and growth media.

After inserting the DNA sequence encoding the protein into the transfer vector, the vector and the wild type viral genome are transfected into an insect host cell where the vector and viral genome are allowed to recombine. The packaged recombinant virus is expressed and recombinant plaques are identified and purified. Materials and methods for baculovirus/insect cell expression systems are commercially available in kit form from, *inter alia*, Invitrogen, San Diego CA ("MaxBac" kit). These techniques are generally known to those skilled in the art and fully described in Summers and Smith, *Texas Agricultural Experiment Station Bulletin No. 1555* (1987) (hereinafter "Summers and Smith").

Prior to inserting the DNA sequence encoding the protein into the baculovirus genome, the above described components, comprising a promoter, leader (if desired), coding sequence of interest, and transcription termination sequence, are usually assembled into an intermediate transplacement construct (transfer vector). This construct may contain a single gene and operably linked regulatory elements; multiple genes, each with its owned set of operably linked regulatory elements; or multiple genes, regulated by the same set of regulatory elements. Intermediate transplacement constructs are often maintained in a replicon, such as an extrachromosomal element (e.g., plasmids) capable of stable maintenance in a host, such as a bacterium. The replicon will have a replication system, thus allowing it to be maintained in a suitable host for cloning and amplification.

Currently, the most commonly used transfer vector for introducing foreign genes into AcNPV is pAc373. Many other vectors, known to those of skill in the art, have also been designed. These include, for example, pVL985 (which alters the polyhedrin start codon from ATG to ATT, and which introduces a BamHI cloning site 32 basepairs downstream from the ATT; see Luckow and Summers, *Virology* (1989) 17:31.

The plasmid usually also contains the polyhedrin polyadenylation signal (Miller et al. (1988) *Ann. Rev. Microbiol.*, 42:177) and a prokaryotic ampicillin-resistance (*amp*) gene and origin of replication for selection and propagation in *E. coli*.

Baculovirus transfer vectors usually contain a baculovirus promoter. A baculovirus promoter is any DNA sequence capable of binding a baculovirus RNA polymerase and initiating the downstream (5' to 3') transcription of a coding sequence (e.g., structural gene)

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into mRNA. A promoter will have a transcription initiation region which is usually placed proximal to the 5' end of the coding sequence. This transcription initiation region usually includes an RNA polymerase binding site and a transcription initiation site. A baculovirus transfer vector may also have a second domain called an enhancer, which, if present, is usually distal to the structural gene. Expression may be either regulated or constitutive.

Structural genes, abundantly transcribed at late times in a viral infection cycle, provide particularly useful promoter sequences. Examples include sequences derived from the gene encoding the viral polyhedron protein, Friesen et al., (1986) "The Regulation of Baculovirus Gene Expression," in: *The Molecular Biology of Baculoviruses* (ed. Walter Doerfler); EPO Publ. Nos. 127 839 and 155 476; and the gene encoding the p10 protein, Vlcek et al., (1988), *J. Gen. Virol.* 69:765.

DNA encoding suitable signal sequences can be derived from genes for secreted insect or baculovirus proteins, such as the baculovirus polyhedrin gene (Carbonell et al. (1988) *Gene*, 73:409). Alternatively, since the signals for mammalian cell posttranslational modifications (such as signal peptide cleavage, proteolytic cleavage, and phosphorylation) appear to be recognized by insect cells, and the signals required for secretion and nuclear accumulation also appear to be conserved between the invertebrate cells and vertebrate cells, leaders of non-insect origin, such as those derived from genes encoding human (alpha) α -interferon, Maeda et al., (1985), *Nature* 315:592; human gastrin-releasing peptide, Lebacqz-Verheyden et al., (1988), *Molec. Cell. Biol.* 8:3129; human IL-2, Smith et al., (1985) *Proc. Nat'l Acad. Sci. USA*, 82:8404; mouse IL-3, (Miyajima et al., (1987) *Gene* 58:273; and human glucocerebrosidase, Martin et al. (1988) *DNA*, 7:99, can also be used to provide for secretion in insects.

A recombinant polypeptide or polyprotein may be expressed intracellularly or, if it is expressed with the proper regulatory sequences, it can be secreted. Good intracellular expression of nonfused foreign proteins usually requires heterologous genes that ideally have a short leader sequence containing suitable translation initiation signals preceding an ATG start signal. If desired, methionine at the N-terminus may be cleaved from the mature protein by *in vitro* incubation with cyanogen bromide.

Alternatively, recombinant polyproteins or proteins which are not naturally secreted can be secreted from the insect cell by creating chimeric DNA molecules that encode a fusion

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protein comprised of a leader sequence fragment that provides for secretion of the foreign protein in insects. The leader sequence fragment usually encodes a signal peptide comprised of hydrophobic amino acids which direct the translocation of the protein into the endoplasmic reticulum.

After insertion of the DNA sequence and/or the gene encoding the expression product precursor of the protein, an insect cell host is co-transformed with the heterologous DNA of the transfer vector and the genomic DNA of wild type baculovirus -- usually by co-transfection. The promoter and transcription termination sequence of the construct will usually comprise a 2-5kb section of the baculovirus genome. Methods for introducing heterologous DNA into the desired site in the baculovirus virus are known in the art. (See Summers and Smith *supra*; Ju et al. (1987); Smith et al., *Mol. Cell. Biol.* (1983) 3:2156; and Luckow and Summers (1989)). For example, the insertion can be into a gene such as the polyhedrin gene, by homologous double crossover recombination; insertion can also be into a restriction enzyme site engineered into the desired baculovirus gene. Miller et al., (1989), *Bioessays* 4:91. The DNA sequence, when cloned in place of the polyhedrin gene in the expression vector, is flanked both 5' and 3' by polyhedrin-specific sequences and is positioned downstream of the polyhedrin promoter.

The newly formed baculovirus expression vector is subsequently packaged into an infectious recombinant baculovirus. Homologous recombination occurs at low frequency (between about 1% and about 5%); thus, the majority of the virus produced after cotransfection is still wild-type virus. Therefore, a method is necessary to identify recombinant viruses. An advantage of the expression system is a visual screen allowing recombinant viruses to be distinguished. The polyhedrin protein, which is produced by the native virus, is produced at very high levels in the nuclei of infected cells at late times after viral infection. Accumulated polyhedrin protein forms occlusion bodies that also contain embedded particles. These occlusion bodies, up to 15 μ m in size, are highly refractile, giving them a bright shiny appearance that is readily visualized under the light microscope. Cells infected with recombinant viruses lack occlusion bodies. To distinguish recombinant virus from wild-type virus, the transfection supernatant is plaqued onto a monolayer of insect cells by techniques known to those skilled in the art. Namely, the plaques are screened under the light microscope for the presence (indicative of wild-type virus) or absence (indicative of

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recombinant virus) of occlusion bodies. *Current Protocols in Microbiology* Vol. 2 (Ausubel et al. eds) at 16.8 (Supp. 10, 1990); Summers and Smith, *supra*; Miller et al. (1989).

Recombinant baculovirus expression vectors have been developed for infection into several insect cells. For example, recombinant baculoviruses have been developed for, *inter alia*: *Aedes aegypti*, *Autographa californica*, *Bombyx mori*, *Drosophila melanogaster*, *Spodoptera frugiperda*, and *Trichoplusia ni* (PCT Pub. No. WO 89/046699; Carbonell et al., (1985) *J. Virol.* 56:153; Wright (1986) *Nature* 321:718; Smith et al., (1983) *Mol. Cell. Biol.* 3:2156; and see generally, Fraser, *et al.* (1989) *In Vitro Cell. Dev. Biol.* 25:225).

Cells and cell culture media are commercially available for both direct and fusion expression of heterologous polypeptides in a baculovirus/expression system; cell culture technology is generally known to those skilled in the art. See, e.g., Summers and Smith *supra*.

The modified insect cells may then be grown in an appropriate nutrient medium, which allows for stable maintenance of the plasmid(s) present in the modified insect host. Where the expression product gene is under inducible control, the host may be grown to high density, and expression induced. Alternatively, where expression is constitutive, the product will be continuously expressed into the medium and the nutrient medium must be continuously circulated, while removing the product of interest and augmenting depleted nutrients. The product may be purified by such techniques as chromatography, e.g., HPLC, affinity chromatography, ion exchange chromatography, etc.; electrophoresis; density gradient centrifugation; solvent extraction, or the like. As appropriate, the product may be further purified, as required, so as to remove substantially any insect proteins which are also secreted in the medium or result from lysis of insect cells, so as to provide a product which is at least substantially free of host debris, e.g., proteins, lipids and polysaccharides.

In order to obtain protein expression, recombinant host cells derived from the transformants are incubated under conditions which allow expression of the recombinant protein encoding sequence. These conditions will vary, dependent upon the host cell selected. However, the conditions are readily ascertainable to those of ordinary skill in the art, based upon what is known in the art.

iv. Bacterial Systems

Bacterial expression techniques are known in the art. A bacterial promoter is any DNA sequence capable of binding bacterial RNA polymerase and initiating the downstream (3') transcription of a coding sequence (e.g. structural gene) into mRNA. A promoter will have a transcription initiation region which is usually placed proximal to the 5' end of the coding sequence. This transcription initiation region usually includes an RNA polymerase binding site and a transcription initiation site. A bacterial promoter may also have a second domain called an operator, that may overlap an adjacent RNA polymerase binding site at which RNA synthesis begins. The operator permits negative regulated (inducible) transcription, as a gene repressor protein may bind the operator and thereby inhibit transcription of a specific gene. Constitutive expression may occur in the absence of negative regulatory elements, such as the operator. In addition, positive regulation may be achieved by a gene activator protein binding sequence, which, if present is usually proximal (5') to the RNA polymerase binding sequence. An example of a gene activator protein is the catabolite activator protein (CAP), which helps initiate transcription of the lac operon in *Escherichia coli* (*E. coli*) (Raibaud *et al.* (1984) *Annu. Rev. Genet.* 18:173). Regulated expression may therefore be either positive or negative, thereby either enhancing or reducing transcription.

Sequences encoding metabolic pathway enzymes provide particularly useful promoter sequences. Examples include promoter sequences derived from sugar metabolizing enzymes, such as galactose, lactose (*lac*) (Chang *et al.* (1977) *Nature* 198:1056), and maltose. Additional examples include promoter sequences derived from biosynthetic enzymes such as tryptophan (*trp*) (Goeddel *et al.* (1980) *Nuc. Acids Res.* 8:4057; Yelverton *et al.* (1981) *Nucl. Acids Res.* 9:731; U.S. Patent 4,738,921; EPO Publ. Nos. 036 776 and 121 775). The beta-lactamase (*bla*) promoter system (Weissmann (1981) "The cloning of interferon and other mistakes." In *Interferon 3* (ed. I. Gresser)), bacteriophage lambda PL (Shimatake *et al.* (1981) *Nature* 292:128) and T5 (U.S. Patent 4,689,406) promoter systems also provide useful promoter sequences.

In addition, synthetic promoters which do not occur in nature also function as bacterial promoters. For example, transcription activation sequences of one bacterial or bacteriophage promoter may be joined with the operon sequences of another bacterial or bacteriophage promoter, creating a synthetic hybrid promoter (U.S. Patent 4,551,433). For

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example, the *tac* promoter is a hybrid *trp-lac* promoter comprised of both *trp* promoter and *lac* operon sequences that is regulated by the *lac* repressor (Amann *et al.* (1983) *Gene* 25:167; de Boer *et al.* (1983) *Proc. Natl. Acad. Sci.* 80:21). Furthermore, a bacterial promoter can include naturally occurring promoters of non-bacterial origin that have the ability to bind bacterial RNA polymerase and initiate transcription. A naturally occurring promoter of non-bacterial origin can also be coupled with a compatible RNA polymerase to produce high levels of expression of some genes in prokaryotes. The bacteriophage T7 RNA polymerase/promoter system is an example of a coupled promoter system (Studier *et al.* (1986) *J. Mol. Biol.* 189:113; Tabor *et al.* (1985) *Proc Natl. Acad. Sci.* 82:1074). In addition, a hybrid promoter can also be comprised of a bacteriophage promoter and an *E. coli* operator region (EPO Publ. No. 267 851).

In addition to a functioning promoter sequence, an efficient ribosome binding site is also useful for the expression of foreign genes in prokaryotes. In *E. coli*, the ribosome binding site is called the Shine-Dalgarno (SD) sequence and includes an initiation codon (ATG) and a sequence 3-9 nucleotides in length located 3-11 nucleotides upstream of the initiation codon (Shine *et al.* (1975) *Nature* 254:34). The SD sequence is thought to promote binding of mRNA to the ribosome by the pairing of bases between the SD sequence and the 3' end of *E. coli* 16S rRNA (Steitz *et al.* (1979) "Genetic signals and nucleotide sequences in messenger RNA." In *Biological Regulation and Development: Gene Expression* (ed. R.F. Goldberg)). To express eukaryotic genes and prokaryotic genes with weak ribosome-binding site, it is often necessary to optimize the distance between the SD sequence and the ATG of the eukaryotic gene (Sambrook *et al.* (1989) "Expression of cloned genes in *Escherichia coli*." In *Molecular Cloning: A Laboratory Manual*).

A DNA molecule may be expressed intracellularly. A promoter sequence may be directly linked with the DNA molecule, in which case the first amino acid at the N-terminus will always be a methionine, which is encoded by the ATG start codon. If desired, methionine at the N-terminus may be cleaved from the protein by *in vitro* incubation with cyanogen bromide or by either *in vivo* or *in vitro* incubation with a bacterial methionine N-terminal peptidase (EPO Publ. No. 219 237).

Fusion proteins provide an alternative to direct expression. Usually, a DNA sequence encoding the N-terminal portion of an endogenous bacterial protein, or other stable protein, is

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fused to the 5' end of heterologous coding sequences. Upon expression, this construct will provide a fusion of the two amino acid sequences. For example, the bacteriophage lambda cell gene can be linked at the 5' terminus of a foreign gene and expressed in bacteria. The resulting fusion protein preferably retains a site for a processing enzyme (factor Xa) to cleave the bacteriophage protein from the foreign gene (Nagai *et al.* (1984) *Nature* 309:810). Fusion proteins can also be made with sequences from the *lacZ* (Jia *et al.* (1987) *Gene* 60:197), *trpE* (Allen *et al.* (1987) *J. Biotechnol.* 5:93; Makoff *et al.* (1989) *J. Gen. Microbiol.* 135:11), and *Chey* (EPO Publ. No. 324 647) genes. The DNA sequence at the junction of the two amino acid sequences may or may not encode a cleavable site. Another example is a ubiquitin fusion protein. Such a fusion protein is made with the ubiquitin region that preferably retains a site for a processing enzyme (e.g. ubiquitin specific processing-protease) to cleave the ubiquitin from the foreign protein. Through this method, native foreign protein can be isolated (Miller *et al.* (1989) *Bio/Technology* 7:698).

Alternatively, foreign proteins can also be secreted from the cell by creating chimeric DNA molecules that encode a fusion protein comprised of a signal peptide sequence fragment that provides for secretion of the foreign protein in bacteria (U.S. Patent 4,336,336). The signal sequence fragment usually encodes a signal peptide comprised of hydrophobic amino acids which direct the secretion of the protein from the cell. The protein is either secreted into the growth media (gram-positive bacteria) or into the periplasmic space, located between the inner and outer membrane of the cell (gram-negative bacteria). Preferably there are processing sites, which can be cleaved either *in vivo* or *in vitro* encoded between the signal peptide fragment and the foreign gene.

DNA encoding suitable signal sequences can be derived from genes for secreted bacterial proteins, such as the *E. coli* outer membrane protein gene (*ompA*) (Masui *et al.* (1983), in: *Experimental Manipulation of Gene Expression*; Ghayeb *et al.* (1984) *EMBO J.* 3:2437) and the *E. coli* alkaline phosphatase signal sequence (*phoA*) (Oka *et al.* (1985) *Proc. Natl. Acad. Sci.* 82:7212). As an additional example, the signal sequence of the alpha-amylase gene from various *Bacillus* strains can be used to secrete heterologous proteins from *B. subtilis* (Palva *et al.* (1982) *Proc. Natl. Acad. Sci. USA* 79:5582; EPO Publ. No. 244 042).

Usually, transcription termination sequences recognized by bacteria are regulatory regions located 3' to the translation stop codon, and thus together with the promoter flank the

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coding sequence. These sequences direct the transcription of an mRNA which can be translated into the polypeptide encoded by the DNA. Transcription termination sequences frequently include DNA sequences of about 50 nucleotides capable of forming stem loop structures that aid in terminating transcription. Examples include transcription termination sequences derived from genes with strong promoters, such as the *trp* gene in *E. coli* as well as other biosynthetic genes.

Usually, the above described components, comprising a promoter, signal sequence (if desired), coding sequence of interest, and transcription termination sequence, are put together into expression constructs. Expression constructs are often maintained in a replicon, such as an extrachromosomal element (e.g., plasmids) capable of stable maintenance in a host, such as bacteria. The replicon will have a replication system, thus allowing it to be maintained in a prokaryotic host either for expression or for cloning and amplification. In addition, a replicon may be either a high or low copy number plasmid. A high copy number plasmid will generally have a copy number ranging from about 5 to about 200, and usually about 10 to about 150. A host containing a high copy number plasmid will preferably contain at least about 10, and more preferably at least about 20 plasmids. Either a high or low copy number vector may be selected, depending upon the effect of the vector and the foreign protein on the host.

Alternatively, the expression constructs can be integrated into the bacterial genome with an integrating vector. Integrating vectors usually contain at least one sequence homologous to the bacterial chromosome that allows the vector to integrate. Integrations appear to result from recombinations between homologous DNA in the vector and the bacterial chromosome. For example, integrating vectors constructed with DNA from various *Bacillus* strains integrate into the *Bacillus* chromosome (EPO Publ. No. 127 328). Integrating vectors may also be comprised of bacteriophage or transposon sequences.

Usually, extrachromosomal and integrating expression constructs may contain selectable markers to allow for the selection of bacterial strains that have been transformed. Selectable markers can be expressed in the bacterial host and may include genes which render bacteria resistant to drugs such as ampicillin, chloramphenicol, erythromycin, kanamycin (neomycin), and tetracycline (Davies *et al.* (1978) *Annu. Rev. Microbiol.* 32:469). Selectable

markers may also include biosynthetic genes, such as those in the histidine, tryptophan, and leucine biosynthetic pathways.

Alternatively, some of the above described components can be put together in transformation vectors. Transformation vectors are usually comprised of a selectable marker that is either maintained in a replicon or developed into an integrating vector, as described above.

Expression and transformation vectors, either extra-chromosomal replicons or integrating vectors, have been developed for transformation into many bacteria. For example, expression vectors have been developed for, *inter alia*, the following bacteria: *Bacillus subtilis* (Palva *et al.* (1982) *Proc. Natl. Acad. Sci. USA* 79:5582; EPO Publ. Nos. 036 259 and 063 953; PCT Publ. No. WO 84/04541), *Escherichia coli* (Shimatake *et al.* (1981) *Nature* 292:128; Amann *et al.* (1985) *Gene* 40:183; Studier *et al.* (1986) *J. Mol. Biol.* 189:113; EPO Publ. Nos. 036 776, 136 829 and 136 907), *Streptococcus cremoris* (Powell *et al.* (1988) *Appl. Environ. Microbiol.* 54:655); *Streptococcus lividans* (Powell *et al.* (1988) *Appl. Environ. Microbiol.* 54:655), *Streptomyces lividans* (U.S. Patent 4,745,056).

Methods of introducing exogenous DNA into bacterial hosts are well-known in the art, and usually include either the transformation of bacteria treated with CaCl_2 or other agents, such as divalent cations and DMSO. DNA can also be introduced into bacterial cells by electroporation. Transformation procedures usually vary with the bacterial species to be transformed. (See e.g., use of *Bacillus*: Masson *et al.* (1989) *FEMS Microbiol. Lett.* 60:273; Palva *et al.* (1982) *Proc. Natl. Acad. Sci. USA* 79:5582; EPO Publ. Nos. 036 259 and 063 953; PCT Publ. No. WO 84/04541; use of *Campylobacter*: Miller *et al.* (1988) *Proc. Natl. Acad. Sci.* 85:856; and Wang *et al.* (1990) *J. Bacteriol.* 172:949; use of *Escherichia coli*: Cohen *et al.* (1973) *Proc. Natl. Acad. Sci.* 69:2110; Dower *et al.* (1988) *Nucleic Acids Res.* 16:6127; Kushner (1978) "An improved method for transformation of *Escherichia coli* with ColE1-derived plasmids. In *Genetic Engineering: Proceedings of the International Symposium on Genetic Engineering* (eds. H.W. Boyer and S. Nicosia); Mandel *et al.* (1970) *J. Mol. Biol.* 53:159; Taketo (1988) *Biochim. Biophys. Acta* 949:318; use of *Lactobacillus*: Chassy *et al.* (1987) *FEMS Microbiol. Lett.* 44:173; use of *Pseudomonas*: Fiedler *et al.* (1988) *Anal. Biochem.* 170:38; use of *Staphylococcus*: Augustin *et al.* (1990) *FEMS Microbiol. Lett.* 66:203; use of *Streptococcus*: Barany *et al.* (1980) *J. Bacteriol.* 144:698;

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Harlander (1987) "Transformation of *Streptococcus lactis* by electroporation, in: *Streptococcal Genetics* (ed. J. Ferretti and R. Curtiss III); Perry *et al.* (1981) *Infect. Immun.* 32:1295; Powell *et al.* (1988) *Appl. Environ. Microbiol.* 54:655; Somkuti *et al.* (1987) *Proc. 4th Evr. Cong. Biotechnology* 1:412.

v. Yeast Expression

Yeast expression systems are also known to one of ordinary skill in the art. A yeast promoter is any DNA sequence capable of binding yeast RNA polymerase and initiating the downstream (3') transcription of a coding sequence (e.g. structural gene) into mRNA. A promoter will have a transcription initiation region which is usually placed proximal to the 5' end of the coding sequence. This transcription initiation region usually includes an RNA polymerase binding site (the "TATA Box") and a transcription initiation site. A yeast promoter may also have a second domain called an upstream activator sequence (UAS), which, if present, is usually distal to the structural gene. The UAS permits regulated (inducible) expression. Constitutive expression occurs in the absence of a UAS. Regulated expression may be either positive or negative, thereby either enhancing or reducing transcription.

Yeast is a fermenting organism with an active metabolic pathway, therefore sequences encoding enzymes in the metabolic pathway provide particularly useful promoter sequences. Examples include alcohol dehydrogenase (ADH) (EPO Publ. No. 284 044), enolase, glucokinase, glucose-6-phosphate isomerase, glyceraldehyde-3-phosphate-dehydrogenase (GAP or GAPDH), hexokinase, phosphofructokinase, 3-phosphoglycerate mutase, and pyruvate kinase (PyK) (EPO Publ. No. 329 203). The yeast *PHO5* gene, encoding acid phosphatase, also provides useful promoter sequences (Myanohara *et al.* (1983) *Proc. Natl. Acad. Sci. USA* 80:1).

In addition, synthetic promoters which do not occur in nature also function as yeast promoters. For example, UAS sequences of one yeast promoter may be joined with the transcription activation region of another yeast promoter, creating a synthetic hybrid promoter. Examples of such hybrid promoters include the ADH regulatory sequence linked to the GAP transcription activation region (U.S. Patent Nos. 4,876,197 and 4,880,734). Other examples of hybrid promoters include promoters which consist of the regulatory sequences of

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either the *ADH2*, *GAL4*, *GAL10*, OR *PHO5* genes, combined with the transcriptional activation region of a glycolytic enzyme gene such as GAP or PyK (EPO Publ. No. 164 556). Furthermore, a yeast promoter can include naturally occurring promoters of non-yeast origin that have the ability to bind yeast RNA polymerase and initiate transcription. Examples of such promoters include, *inter alia*, (Cohen *et al.* (1980) *Proc. Natl. Acad. Sci. USA* 77:1078; Henikoff *et al.* (1981) *Nature* 283:835; Hollenberg *et al.* (1981) *Curr. Topics Microbiol. Immunol.* 96:119; Hollenberg *et al.* (1979) "The Expression of Bacterial Antibiotic Resistance Genes in the Yeast *Saccharomyces cerevisiae*," in: *Plasmids of Medical, Environmental and Commercial Importance* (eds. K.N. Timmis and A. Puhler); Mercerau-Puigalon *et al.* (1980) *Gene* 11:163; Panthier *et al.* (1980) *Curr. Genet.* 2:109;).

A DNA molecule may be expressed intracellularly in yeast. A promoter sequence may be directly linked with the DNA molecule, in which case the first amino acid at the N-terminus of the recombinant protein will always be a methionine, which is encoded by the ATG start codon. If desired, methionine at the N-terminus may be cleaved from the protein by *in vitro* incubation with cyanogen bromide.

Fusion proteins provide an alternative for yeast expression systems, as well as in mammalian, plant, baculovirus, and bacterial expression systems. Usually, a DNA sequence encoding the N-terminal portion of an endogenous yeast protein, or other stable protein, is fused to the 5' end of heterologous coding sequences. Upon expression, this construct will provide a fusion of the two amino acid sequences. For example, the yeast or human superoxide dismutase (SOD) gene, can be linked at the 5' terminus of a foreign gene and expressed in yeast. The DNA sequence at the junction of the two amino acid sequences may or may not encode a cleavable site. See e.g., EPO Publ. No. 196056. Another example is a ubiquitin fusion protein. Such a fusion protein is made with the ubiquitin region that preferably retains a site for a processing enzyme (e.g. ubiquitin-specific processing protease) to cleave the ubiquitin from the foreign protein. Through this method, therefore, native foreign protein can be isolated (e.g., WO88/024066).

Alternatively, foreign proteins can also be secreted from the cell into the growth media by creating chimeric DNA molecules that encode a fusion protein comprised of a leader sequence fragment that provide for secretion in yeast of the foreign protein. Preferably, there are processing sites encoded between the leader fragment and the foreign gene that can

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be cleaved either *in vivo* or *in vitro*. The leader sequence fragment usually encodes a signal peptide comprised of hydrophobic amino acids which direct the secretion of the protein from the cell.

DNA encoding suitable signal sequences can be derived from genes for secreted yeast proteins, such as the yeast invertase gene (EPO Publ. No. 012 873; JPO Publ. No. 62:096,086) and the A-factor gene (U.S. Patent 4,588,684). Alternatively, leaders of non-yeast origin, such as an interferon leader, exist that also provide for secretion in yeast (EPO Publ. No. 060 057).

A preferred class of secretion leaders are those that employ a fragment of the yeast alpha-factor gene, which contains both a "pre" signal sequence, and a "pro" region. The types of alpha-factor fragments that can be employed include the full-length pre-pro alpha factor leader (about 83 amino acid residues) as well as truncated alpha-factor leaders (usually about 25 to about 50 amino acid residues) (U.S. Patent Nos. 4,546,083 and 4,870,008; EPO Publ. No. 324 274). Additional leaders employing an alpha-factor leader fragment that provides for secretion include hybrid alpha-factor leaders made with a presequence of a first yeast, but a pro-region from a second yeast alpha factor. (See e.g., PCT Publ. No. WO 89/02463.)

Usually, transcription termination sequences recognized by yeast are regulatory regions located 3' to the translation stop codon, and thus together with the promoter flank the coding sequence. These sequences direct the transcription of an mRNA which can be translated into the polypeptide encoded by the DNA. Examples of transcription terminator sequence and other yeast-recognized termination sequences, such as those coding for glycolytic enzymes.

Usually, the above described components, comprising a promoter, leader (if desired), coding sequence of interest, and transcription termination sequence, are put together into expression constructs. Expression constructs are often maintained in a replicon, such as an extrachromosomal element (e.g., plasmids) capable of stable maintenance in a host, such as yeast or bacteria. The replicon may have two replication systems, thus allowing it to be maintained, for example, in yeast for expression and in a prokaryotic host for cloning and amplification. Examples of such yeast-bacteria shuttle vectors include YEp24 (Botstein *et al.* (1979) *Gene* 8:17-24), pCl/1 (Brake *et al.* (1984) *Proc. Natl. Acad. Sci USA* 81:4642-4646), and YRp17 (Stinchcomb *et al.* (1982) *J. Mol. Biol.* 158:157). In addition, a replicon may be

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either a high or low copy number plasmid. A high copy number plasmid will generally have a copy number ranging from about 5 to about 200, and usually about 10 to about 150. A host containing a high copy number plasmid will preferably have at least about 10, and more preferably at least about 20. Either a high or low copy number vector may be selected, depending upon the effect of the vector and the foreign protein on the host. See e.g., Brake *et al.*, *supra*.

Alternatively, the expression constructs can be integrated into the yeast genome with an integrating vector. Integrating vectors usually contain at least one sequence homologous to a yeast chromosome that allows the vector to integrate, and preferably contain two homologous sequences flanking the expression construct. Integrations appear to result from recombinations between homologous DNA in the vector and the yeast chromosome (Orr-Weaver *et al.* (1983) *Methods in Enzymol.* 101:228-245). An integrating vector may be directed to a specific locus in yeast by selecting the appropriate homologous sequence for inclusion in the vector. See Orr-Weaver *et al.*, *supra*. One or more expression construct may integrate, possibly affecting levels of recombinant protein produced (Rine *et al.* (1983) *Proc. Natl. Acad. Sci. USA* 80:6750). The chromosomal sequences included in the vector can occur either as a single segment in the vector, which results in the integration of the entire vector, or two segments homologous to adjacent segments in the chromosome and flanking the expression construct in the vector, which can result in the stable integration of only the expression construct.

Usually, extrachromosomal and integrating expression constructs may contain selectable markers to allow for the selection of yeast strains that have been transformed. Selectable markers may include biosynthetic genes that can be expressed in the yeast host, such as *ADE2*, *HIS4*, *LEU2*, *TRP1*, and *ALG7*, and the G418 resistance gene, which confer resistance in yeast cells to tunicamycin and G418, respectively. In addition, a suitable selectable marker may also provide yeast with the ability to grow in the presence of toxic compounds, such as metal. For example, the presence of *CUP1* allows yeast to grow in the presence of copper ions (Butt *et al.* (1987) *Microbiol. Rev.* 51:351).

Alternatively, some of the above described components can be put together into transformation vectors. Transformation vectors are usually comprised of a selectable marker

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that is either maintained in a replicon or developed into an integrating vector, as described above.

Expression and transformation vectors, either extrachromosomal replicons or integrating vectors, have been developed for transformation into many yeasts. For example, expression vectors and methods of introducing exogenous DNA into yeast hosts have been developed for, *inter alia*, the following yeasts: *Candida albicans* (Kurtz, *et al.* (1986) *Mol. Cell. Biol.* 6:142); *Candida maltosa* (Kunze, *et al.* (1985) *J. Basic Microbiol.* 25:141); *Hansenula polymorpha* (Gleeson, *et al.* (1986) *J. Gen. Microbiol.* 132:3459; Roggenkamp *et al.* (1986) *Mol. Gen. Genet.* 202:302); *Kluyveromyces fragilis* (Das, *et al.* (1984) *J. Bacteriol.* 158:1165); *Kluyveromyces lactis* (De Louvencourt *et al.* (1983) *J. Bacteriol.* 154:737; Van den Berg *et al.* (1990) *Bio/Technology* 8:135); *Pichia guillermondii* (Kunze *et al.* (1985) *J. Basic Microbiol.* 25:141); *Pichia pastoris* (Cregg, *et al.* (1985) *Mol. Cell. Biol.* 5:3376; U.S. Patent Nos. 4,837,148 and 4,929,555); *Saccharomyces cerevisiae* (Hinnen *et al.* (1978) *Proc. Natl. Acad. Sci. USA* 75:1929; Ito *et al.* (1983) *J. Bacteriol.* 153:163); *Schizosaccharomyces pombe* (Beach and Nurse (1981) *Nature* 300:706); and *Yarrowia lipolytica* (Davidow, *et al.* (1985) *Curr. Genet.* 10:380471 Gaillardin, *et al.* (1985) *Curr. Genet.* 10:49).

Methods of introducing exogenous DNA into yeast hosts are well-known in the art, and usually include either the transformation of spheroplasts or of intact yeast cells treated with alkali cations. Transformation procedures usually vary with the yeast species to be transformed. See e.g., [Kurtz *et al.* (1986) *Mol. Cell. Biol.* 6:142; Kunze *et al.* (1985) *J. Basic Microbiol.* 25:141; *Candida*]; [Gleeson *et al.* (1986) *J. Gen. Microbiol.* 132:3459; Roggenkamp *et al.* (1986) *Mol. Gen. Genet.* 202:302; *Hansenula*]; [Das *et al.* (1984) *J. Bacteriol.* 158:1165; De Louvencourt *et al.* (1983) *J. Bacteriol.* 154:1165; Van den Berg *et al.* (1990) *Bio/Technology* 8:135; *Kluyveromyces*]; [Cregg *et al.* (1985) *Mol. Cell. Biol.* 5:3376; Kunze *et al.* (1985) *J. Basic Microbiol.* 25:141; U.S. Patent Nos. 4,837,148 and 4,929,555; *Pichia*]; [Hinnen *et al.* (1978) *Proc. Natl. Acad. Sci. USA* 75:1929; Ito *et al.* (1983) *J. Bacteriol.* 153:163 *Saccharomyces*]; [Beach and Nurse (1981) *Nature* 300:706; *Schizosaccharomyces*]; [Davidow *et al.* (1985) *Curr. Genet.* 10:39; Gaillardin *et al.* (1985) *Curr. Genet.* 10:49; *Yarrowia*].

Definitions

A composition containing X is “substantially free of” Y when at least 85% by weight of the total X+Y in the composition is X. Preferably, X comprises at least about 90% by weight of the total of X+Y in the composition, more preferably at least about 95% or even 99% by weight.

The term “heterologous” refers to two biological components that are not found together in nature. The components may be host cells, genes, or regulatory regions, such as promoters. Although the heterologous components are not found together in nature, they can function together, as when a promoter heterologous to a gene is operably linked to the gene. Another example is where a Neisserial sequence is heterologous to a mouse host cell.

An “origin of replication” is a polynucleotide sequence that initiates and regulates replication of polynucleotides, such as an expression vector. The origin of replication behaves as an autonomous unit of polynucleotide replication within a cell, capable of replication under its own control. An origin of replication may be needed for a vector to replicate in a particular host cell. With certain origins of replication, an expression vector can be reproduced at a high copy number in the presence of the appropriate proteins within the cell. Examples of origins are the autonomously replicating sequences, which are effective in yeast; and the viral T-antigen, effective in COS-7 cells.

A “mutant” sequence is defined as a DNA, RNA or amino acid sequence differing from but having homology with the native or disclosed sequence. Depending on the particular sequence, the degree of homology between the native or disclosed sequence and the mutant sequence is preferably greater than 50% (e.g., 60%, 70%, 80%, 90%, 95%, 99% or more) which is calculated as described above. As used herein, an “allelic variant” of a nucleic acid molecule, or region, for which nucleic acid sequence is provided herein is a nucleic acid molecule, or region, that occurs at essentially the same locus in the genome of another or second isolate, and that, due to natural variation caused by, for example, mutation or recombination, has a similar but not identical nucleic acid sequence. A coding region allelic variant typically encodes a protein having similar activity to that of the protein encoded by the gene to which it is being compared. An allelic variant can also comprise an alteration in the 5' or 3' untranslated regions of the gene, such as in regulatory control regions. (see, for example, U.S. Patent 5,753,235).

Antibodies

As used herein, the term “antibody” refers to a polypeptide or group of polypeptides composed of at least one antibody combining site. An “antibody combining site” is the three-dimensional binding space with an internal surface shape and charge distribution complementary to the features of an epitope of an antigen, which allows a binding of the antibody with the antigen. “Antibody” includes, for example, vertebrate antibodies, hybrid antibodies, chimeric antibodies, humanized antibodies, altered antibodies, univalent antibodies, Fab proteins, and single domain antibodies.

Antibodies against the proteins of the invention are useful for affinity chromatography, immunoassays, and distinguishing/identifying *Neisseria* MenB proteins. Antibodies elicited against the proteins of the present invention bind to antigenic polypeptides or proteins or protein fragments that are present and specifically associated with strains of *Neisseria meningitidis* MenB. In some instances, these antigens may be associated with specific strains, such as those antigens specific for the MenB strains. The antibodies of the invention may be immobilized to a matrix and utilized in an immunoassay or on an affinity chromatography column, to enable the detection and/or separation of polypeptides, proteins or protein fragments or cells comprising such polypeptides, proteins or protein fragments. Alternatively, such polypeptides, proteins or protein fragments may be immobilized so as to detect antibodies bindably specific thereto.

Antibodies to the proteins of the invention, both polyclonal and monoclonal, may be prepared by conventional methods. In general, the protein is first used to immunize a suitable animal, preferably a mouse, rat, rabbit or goat. Rabbits and goats are preferred for the preparation of polyclonal sera due to the volume of serum obtainable, and the availability of labeled anti-rabbit and anti-goat antibodies. Immunization is generally performed by mixing or emulsifying the protein in saline, preferably in an adjuvant such as Freund's complete adjuvant, and injecting the mixture or emulsion parenterally (generally subcutaneously or intramuscularly). A dose of 50-200 µg/injection is typically sufficient. Immunization is generally boosted 2-6 weeks later with one or more injections of the protein in saline, preferably using Freund's incomplete adjuvant. One may alternatively generate antibodies by in vitro immunization using methods known in the art, which for the purposes of this

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invention is considered equivalent to *in vivo* immunization. Polyclonal antisera is obtained by bleeding the immunized animal into a glass or plastic container, incubating the blood at 25°C for one hour, followed by incubating at 4°C for 2-18 hours. The serum is recovered by centrifugation (e.g., 1,000g for 10 minutes). About 20-50 ml per bleed may be obtained from rabbits.

Monoclonal antibodies are prepared using the standard method of Kohler & Milstein (*Nature* (1975) 256:495-96), or a modification thereof. Typically, a mouse or rat is immunized as described above. However, rather than bleeding the animal to extract serum, the spleen (and optionally several large lymph nodes) is removed and dissociated into single cells. If desired, the spleen cells may be screened (after removal of nonspecifically adherent cells) by applying a cell suspension to a plate or well coated with the protein antigen. B-cells that express membrane-bound immunoglobulin specific for the antigen bind to the plate, and are not rinsed away with the rest of the suspension. Resulting B-cells, or all dissociated spleen cells, are then induced to fuse with myeloma cells to form hybridomas, and are cultured in a selective medium (e.g., hypoxanthine, aminopterin, thymidine medium, "HAT"). The resulting hybridomas are plated by limiting dilution, and are assayed for the production of antibodies which bind specifically to the immunizing antigen (and which do not bind to unrelated antigens). The selected MAb-secreting hybridomas are then cultured either *in vitro* (e.g., in tissue culture bottles or hollow fiber reactors), or *in vivo* (as ascites in mice).

If desired, the antibodies (whether polyclonal or monoclonal) may be labeled using conventional techniques. Suitable labels include fluorophores, chromophores, radioactive atoms (particularly ³²P and ¹²⁵I), electron-dense reagents, enzymes, and ligands having specific binding partners. Enzymes are typically detected by their activity. For example, horseradish peroxidase is usually detected by its ability to convert 3,3',5,5'-tetramethylbenzidine (TMB) to a blue pigment, quantifiable with a spectrophotometer. "Specific binding partner" refers to a protein capable of binding a ligand molecule with high specificity, as for example in the case of an antigen and a monoclonal antibody specific therefor. Other specific binding partners include biotin and avidin or streptavidin, IgG and protein A, and the numerous receptor-ligand couples known in the art. It should be understood that the above description is not meant to categorize the various

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labels into distinct classes, as the same label may serve in several different modes. For example, ^{125}I may serve as a radioactive label or as an electron-dense reagent. HRP may serve as enzyme or as antigen for a MAb. Further, one may combine various labels for desired effect. For example, MAbs and avidin also require labels in the practice of this invention: thus, one might label a MAb with biotin, and detect its presence with avidin labeled with ^{125}I , or with an anti-biotin MAb labeled with HRP. Other permutations and possibilities will be readily apparent to those of ordinary skill in the art, and are considered as equivalents within the scope of the instant invention.

Antigens, immunogens, polypeptides, proteins or protein fragments of the present invention elicit formation of specific binding partner antibodies. These antigens, immunogens, polypeptides, proteins or protein fragments of the present invention comprise immunogenic compositions of the present invention. Such immunogenic compositions may further comprise or include adjuvants, carriers, or other compositions that promote or enhance or stabilize the antigens, polypeptides, proteins or protein fragments of the present invention. Such adjuvants and carriers will be readily apparent to those of ordinary skill in the art.

Pharmaceutical Compositions

Pharmaceutical compositions can include either polypeptides, antibodies, or nucleic acid of the invention. The pharmaceutical compositions will comprise a therapeutically effective amount of either polypeptides, antibodies, or polynucleotides of the claimed invention.

The term "therapeutically effective amount" as used herein refers to an amount of a therapeutic agent to treat, ameliorate, or prevent a desired disease or condition, or to exhibit a detectable therapeutic or preventative effect. The effect can be detected by, for example, chemical markers or antigen levels. Therapeutic effects also include reduction in physical symptoms, such as decreased body temperature, when given to a patient that is febrile. The precise effective amount for a subject will depend upon the subject's size and health, the nature and extent of the condition, and the therapeutics or combination of therapeutics selected for administration. Thus, it is not useful to specify an exact effective amount in

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advance. However, the effective amount for a given situation can be determined by routine experimentation and is within the judgment of the clinician.

For purposes of the present invention, an effective dose will be from about 0.01 mg/kg to 50 mg/kg or 0.05 mg/kg to about 10 mg/kg of the DNA constructs in the individual to which it is administered.

A pharmaceutical composition can also contain a pharmaceutically acceptable carrier. The term "pharmaceutically acceptable carrier" refers to a carrier for administration of a therapeutic agent, such as antibodies or a polypeptide, genes, and other therapeutic agents. The term refers to any pharmaceutical carrier that does not itself induce the production of antibodies harmful to the individual receiving the composition, and which may be administered without undue toxicity. Suitable carriers may be large, slowly metabolized macromolecules such as proteins, polysaccharides, polylactic acids, polyglycolic acids, polymeric amino acids, amino acid copolymers, and inactive virus particles. Such carriers are well known to those of ordinary skill in the art.

Pharmaceutically acceptable salts can be used therein, for example, mineral acid salts such as hydrochlorides, hydrobromides, phosphates, sulfates, and the like; and the salts of organic acids such as acetates, propionates, malonates, benzoates, and the like. A thorough discussion of pharmaceutically acceptable excipients is available in Remington's Pharmaceutical Sciences (Mack Pub. Co., N.J. 1991).

Pharmaceutically acceptable carriers in therapeutic compositions may contain liquids such as water, saline, glycerol and ethanol. Additionally, auxiliary substances, such as wetting or emulsifying agents, pH buffering substances, and the like, may be present in such vehicles. Typically, the therapeutic compositions are prepared as injectables, either as liquid solutions or suspensions; solid forms suitable for solution in, or suspension in, liquid vehicles prior to injection may also be prepared. Liposomes are included within the definition of a pharmaceutically acceptable carrier.

Delivery Methods

Once formulated, the compositions of the invention can be administered directly to the subject. The subjects to be treated can be animals; in particular, human subjects can be treated.

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Direct delivery of the compositions will generally be accomplished by injection, either subcutaneously, intraperitoneally, intravenously or intramuscularly or delivered to the interstitial space of a tissue. The compositions can also be administered into a lesion. Other modes of administration include oral and pulmonary administration, suppositories, and transdermal and transcutaneous applications, needles, and gene guns or hyposprays. Dosage treatment may be a single dose schedule or a multiple dose schedule.

Vaccines

Vaccines according to the invention may either be prophylactic (i.e., to prevent infection) or therapeutic (i.e., to treat disease after infection).

Such vaccines comprise immunizing antigen(s) or immunogen(s), immunogenic polypeptide, protein(s) or protein fragments, or nucleic acids (e.g., ribonucleic acid or deoxyribonucleic acid), usually in combination with "pharmaceutically acceptable carriers," which include any carrier that does not itself induce the production of antibodies harmful to the individual receiving the composition. Suitable carriers are typically large, slowly metabolized macromolecules such as proteins, polysaccharides, polylactic acids, polyglycolic acids, polymeric amino acids, amino acid copolymers, lipid aggregates (such as oil droplets or liposomes), and inactive virus particles. Such carriers are well known to those of ordinary skill in the art. Additionally, these carriers may function as immunostimulating agents ("adjuvants"). Furthermore, the immunogen or antigen may be conjugated to a bacterial toxoid, such as a toxoid from diphtheria, tetanus, cholera, *H. pylori*, etc. pathogens.

Preferred adjuvants to enhance effectiveness of the composition include, but are not limited to: (1) aluminum salts (alum), such as aluminum hydroxide, aluminum phosphate, aluminum sulfate, etc; (2) oil-in-water emulsion formulations (with or without other specific immunostimulating agents such as muramyl peptides (see below) or bacterial cell wall components), such as for example (a) MF59 (PCT Publ. No. WO 90/14837), containing 5% Squalene, 0.5% Tween 80, and 0.5% Span 85 (optionally containing various amounts of MTP-PE (see below), although not required) formulated into submicron particles using a microfluidizer such as Model 110Y microfluidizer (Microfluidics, Newton, MA), (b) SAF, containing 10% Squalene, 0.4% Tween 80, 5% pluronic-blocked polymer L121, and thr-MDP (see below) either microfluidized into a submicron emulsion or vortexed to generate a

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larger particle size emulsion, and (c) RibiTM adjuvant system (RAS), (Ribi Immunochem, Hamilton, MT) containing 2% Squalene, 0.2% Tween 80, and one or more bacterial cell wall components from the group consisting of monophosphorylipid A (MPL), trehalose dimycolate (TDM), and cell wall skeleton (CWS), preferably MPL + CWS (DetoxTM); (3) saponin adjuvants, such as StimulonTM (Cambridge Bioscience, Worcester, MA) may be used or particles generated therefrom such as ISCOMs (immunostimulating complexes); (4) Complete Freund's Adjuvant (CFA) and Incomplete Freund's Adjuvant (IFA); (5) cytokines, such as interleukins (e.g., IL-1, IL-2, IL-4, IL-5, IL-6, IL-7, IL-12, *etc.*), interferons (e.g., gamma interferon), macrophage colony stimulating factor (M-CSF), tumor necrosis factor (TNF), *etc.*; (6) detoxified mutants of a bacterial ADP-ribosylating toxin such as a cholera toxin (CT), a pertussis toxin (PT), or an *E. coli* heat-labile toxin (LT), particularly LT-K63, LT-R72, CT-S109, PT-K9/G129; see, e.g., WO 93/13302 and WO 92/19265; and (7) other substances that act as immunostimulating agents to enhance the effectiveness of the composition. Alum and MF59 are preferred.

As mentioned above, muramyl peptides include, but are not limited to, N-acetyl-muramyl-L-threonyl-D-isoglutamine (thr-MDP), N-acetyl-normuramyl-L-alanyl-D-isoglutamine (nor-MDP), N-acetylmuramyl-L-alanyl-D-isoglutaminyl-L-alanine-2-(1'-2'-dipalmitoyl-*sn*-glycero-3-hydroxyphosphoryloxy)-ethylamine (MTP-PE), *etc.*

The vaccine compositions comprising immunogenic compositions (e.g., which may include the antigen, pharmaceutically acceptable carrier, and adjuvant) typically will contain diluents, such as water, saline, glycerol, ethanol, *etc.* Additionally, auxiliary substances, such as wetting or emulsifying agents, pH buffering substances, and the like, may be present in such vehicles. Alternatively, vaccine compositions comprising immunogenic compositions may comprise an antigen, polypeptide, protein, protein fragment or nucleic acid in a pharmaceutically acceptable carrier.

More specifically, vaccines comprising immunogenic compositions comprise an immunologically effective amount of the immunogenic polypeptides, as well as any other of the above-mentioned components, as needed. By "immunologically effective amount", it is meant that the administration of that amount to an individual, either in a single dose or as part of a series, is effective for treatment or prevention. This amount varies depending upon the health and physical condition of the individual to be treated, the taxonomic group of

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individual to be treated (e.g., nonhuman primate, primate, *etc.*), the capacity of the individual's immune system to synthesize antibodies, the degree of protection desired, the formulation of the vaccine, the treating doctor's assessment of the medical situation, and other relevant factors. It is expected that the amount will fall in a relatively broad range that can be determined through routine trials.

Typically, the vaccine compositions or immunogenic compositions are prepared as injectables, either as liquid solutions or suspensions; solid forms suitable for solution in, or suspension in, liquid vehicles prior to injection may also be prepared. The preparation also may be emulsified or encapsulated in liposomes for enhanced adjuvant effect, as discussed above under pharmaceutically acceptable carriers.

The immunogenic compositions are conventionally administered parenterally, e.g., by injection, either subcutaneously or intramuscularly. Additional formulations suitable for other modes of administration include oral and pulmonary formulations, suppositories, and transdermal and transcutaneous applications. Dosage treatment may be a single dose schedule or a multiple dose schedule. The vaccine may be administered in conjunction with other immunoregulatory agents.

As an alternative to protein-based vaccines, DNA vaccination may be employed (e.g., Robinson & Torres (1997) *Seminars in Immunology* 9:271-283; Donnelly *et al.* (1997) *Annu Rev Immunol* 15:617-648).

Gene Delivery Vehicles

Gene therapy vehicles for delivery of constructs, including a coding sequence of a therapeutic of the invention, to be delivered to the mammal for expression in the mammal, can be administered either locally or systemically. These constructs can utilize viral or non-viral vector approaches in *in vivo* or *ex vivo* modality. Expression of such coding sequence can be induced using endogenous mammalian or heterologous promoters. Expression of the coding sequence *in vivo* can be either constitutive or regulated.

The invention includes gene delivery vehicles capable of expressing the contemplated nucleic acid sequences. The gene delivery vehicle is preferably a viral vector and, more preferably, a retroviral, adenoviral, adeno-associated viral (AAV), herpes viral, or alphavirus vector. The viral vector can also be an astrovirus, coronavirus, orthomyxovirus, papovavirus,

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paramyxovirus, parvovirus, picornavirus, poxvirus, or togavirus viral vector. See generally, Jolly (1994) *Cancer Gene Therapy* 1:51-64; Kimura (1994) *Human Gene Therapy* 5:845-852; Connelly (1995) *Human Gene Therapy* 6:185-193; and Kaplitt (1994) *Nature Genetics* 6:148-153.

Retroviral vectors are well known in the art, including B, C and D type retroviruses, xenotropic retroviruses (for example, NZB-X1, NZB-X2 and NZB9-1 (see O'Neill (1985) *J. Virol.* 53:160) polytropic retroviruses e.g., MCF and MCF-MLV (see Kelly (1983) *J. Virol.* 45:291), spumaviruses and lentiviruses. See RNA Tumor Viruses, Second Edition, Cold Spring Harbor Laboratory, 1985.

Portions of the retroviral gene therapy vector may be derived from different retroviruses. For example, retrovector LTRs may be derived from a Murine Sarcoma Virus, a tRNA binding site from a Rous Sarcoma Virus, a packaging signal from a Murine Leukemia Virus, and an origin of second strand synthesis from an Avian Leukosis Virus.

These recombinant retroviral vectors may be used to generate transduction competent retroviral vector particles by introducing them into appropriate packaging cell lines (see US patent 5,591,624). Retrovirus vectors can be constructed for site-specific integration into host cell DNA by incorporation of a chimeric integrase enzyme into the retroviral particle (see WO96/37626). It is preferable that the recombinant viral vector is a replication defective recombinant virus.

Packaging cell lines suitable for use with the above-described retrovirus vectors are well known in the art, are readily prepared (see WO95/30763 and WO92/05266), and can be used to create producer cell lines (also termed vector cell lines or "VCLs") for the production of recombinant vector particles. Preferably, the packaging cell lines are made from human parent cells (e.g., HT1080 cells) or mink parent cell lines, which eliminates inactivation in human serum.

Preferred retroviruses for the construction of retroviral gene therapy vectors include Avian Leukosis Virus, Bovine Leukemia, Virus, Murine Leukemia Virus, Mink-Cell Focus-Inducing Virus, Murine Sarcoma Virus, Reticuloendotheliosis Virus and Rous Sarcoma Virus. Particularly preferred Murine Leukemia Viruses include 4070A and 1504A (Hartley and Rowe (1976) *J Virol* 19:19-25), Abelson (ATCC No. VR-999), Friend (ATCC No. VR-245), Graffi, Gross (ATCC No1 VR-590), Kirsten, Harvey Sarcoma Virus and

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Rauscher (ATCC No. VR-998) and Moloney Murine Leukemia Virus (ATCC No. VR-190). Such retroviruses may be obtained from depositories or collections such as the American Type Culture Collection ("ATCC") in Rockville, Maryland or isolated from known sources using commonly available techniques.

Exemplary known retroviral gene therapy vectors employable in this invention include those described in patent applications GB2200651, EP0415731, EP0345242, EP0334301, WO89/02468; WO89/05349, WO89/09271, WO90/02806, WO90/07936, WO94/03622, WO93/25698, WO93/25234, WO93/11230, WO93/10218, WO91/02805, WO91/02825, WO95/07994, US 5,219,740, US 4,405,712, US 4,861,719, US 4,980,289, US 4,777,127, US 5,591,624. See also Vile (1993) *Cancer Res* 53:3860-3864; Vile (1993) *Cancer Res* 53:962-967; Ram (1993) *Cancer Res* 53 (1993) 83-88; Takamiya (1992) *J Neurosci Res* 33:493-503; Baba (1993) *J Neurosurg* 79:729-735; Mann (1983) *Cell* 33:153; Cane (1984) *Proc Natl Acad Sci* 81:6349; and Miller (1990) *Human Gene Therapy* 1.

Human adenoviral gene therapy vectors are also known in the art and employable in this invention. See, for example, Berkner (1988) *Biotechniques* 6:616 and Rosenfeld (1991) *Science* 252:431, and WO93/07283, WO93/06223, and WO93/07282. Exemplary known adenoviral gene therapy vectors employable in this invention include those described in the above referenced documents and in WO94/12649, WO93/03769, WO93/19191, WO94/28938, WO95/11984, WO95/00655, WO95/27071, WO95/29993, WO95/34671, WO96/05320, WO94/08026, WO94/11506, WO93/06223, WO94/24299, WO95/14102, WO95/24297, WO95/02697, WO94/28152, WO94/24299, WO95/09241, WO95/25807, WO95/05835, WO94/18922 and WO95/09654. Alternatively, administration of DNA linked to killed adenovirus as described in Curiel (1992) *Hum. Gene Ther.* 3:147-154 may be employed. The gene delivery vehicles of the invention also include adenovirus associated virus (AAV) vectors. Leading and preferred examples of such vectors for use in this invention are the AAV-2 based vectors disclosed in Srivastava, WO93/09239. Most preferred AAV vectors comprise the two AAV inverted terminal repeats in which the native D-sequences are modified by substitution of nucleotides, such that at least 5 native nucleotides and up to 18 native nucleotides, preferably at least 10 native nucleotides up to 18 native nucleotides, most preferably 10 native nucleotides are retained and the remaining nucleotides of the D-sequence are deleted or replaced with non-native nucleotides. The native

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D-sequences of the AAV inverted terminal repeats are sequences of 20 consecutive nucleotides in each AAV inverted terminal repeat (i.e., there is one sequence at each end) which are not involved in HP formation. The non-native replacement nucleotide may be any nucleotide other than the nucleotide found in the native D-sequence in the same position. Other employable exemplary AAV vectors are pWP-19, pWN-1, both of which are disclosed in Nahreini (1993) *Gene* 124:257-262. Another example of such an AAV vector is psub201 (see Samulski (1987) *J. Virol.* 61:3096). Another exemplary AAV vector is the Double-D ITR vector. Construction of the Double-D ITR vector is disclosed in US Patent 5,478,745. Still other vectors are those disclosed in Carter US Patent 4,797,368 and Muzyczka US Patent 5,139,941, Chartejee US Patent 5,474,935, and Kotin WO94/288157. Yet a further example of an AAV vector employable in this invention is SSV9AFABTKneo, which contains the AFP enhancer and albumin promoter and directs expression predominantly in the liver. Its structure and construction are disclosed in Su (1996) *Human Gene Therapy* 7:463-470. Additional AAV gene therapy vectors are described in US 5,354,678, US 5,173,414, US 5,139,941, and US 5,252,479.

The gene therapy vectors comprising sequences of the invention also include herpes vectors. Leading and preferred examples are herpes simplex virus vectors containing a sequence encoding a thymidine kinase polypeptide such as those disclosed in US 5,288,641 and EP0176170 (Roizman). Additional exemplary herpes simplex virus vectors include HFEM/ICP6-LacZ disclosed in WO95/04139 (Wistar Institute), pHSVlac described in Geller (1988) *Science* 241:1667-1669 and in WO90/09441 and WO92/07945, HSV Us3::pgC-lacZ described in Fink (1992) *Human Gene Therapy* 3:11-19 and HSV 7134, 2 RH 105 and GAL4 described in EP 0453242 (Breakefield), and those deposited with the ATCC as accession numbers ATCC VR-977 and ATCC VR-260.

Also contemplated are alpha virus gene therapy vectors that can be employed in this invention. Preferred alpha virus vectors are Sindbis viruses vectors. Togaviruses, Semliki Forest virus (ATCC VR-67; ATCC VR-1247), Middleberg virus (ATCC VR-370), Ross River virus (ATCC VR-373; ATCC VR-1246), Venezuelan equine encephalitis virus (ATCC VR923; ATCC VR-1250; ATCC VR-1249; ATCC VR-532), and those described in US patents 5,091,309, 5,217,879, and WO92/10578. More particularly, those alpha virus vectors described in U.S. Serial No. 08/405,627, filed March 15, 1995, WO94/21792, WO92/10578,

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WO95/07994, US 5,091,309 and US 5,217,879 are employable. Such alpha viruses may be obtained from depositories or collections such as the ATCC in Rockville, Maryland or isolated from known sources using commonly available techniques. Preferably, alphavirus vectors with reduced cytotoxicity are used (see USSN 08/679640).

DNA vector systems such as eukaryotic layered expression systems are also useful for expressing the nucleic acids of the invention. See WO95/07994 for a detailed description of eukaryotic layered expression systems. Preferably, the eukaryotic layered expression systems of the invention are derived from alphavirus vectors and most preferably from Sindbis viral vectors.

Other viral vectors suitable for use in the present invention include those derived from poliovirus, for example ATCC VR-58 and those described in Evans, *Nature* 339 (1989) 385 and Sabin (1973) *J. Biol. Standardization* 1:115; rhinovirus, for example ATCC VR-1110 and those described in Arnold (1990) *J Cell Biochem* L401; pox viruses such as canary pox virus or vaccinia virus, for example ATCC VR-111 and ATCC VR-2010 and those described in Fisher-Hoch (1989) *Proc Natl Acad Sci* 86:317; Flexner (1989) *Ann NY Acad Sci* 569:86, Flexner (1990) *Vaccine* 8:17; in US 4,603,112 and US 4,769,330 and WO89/01973; SV40 virus, for example ATCC VR-305 and those described in Mulligan (1979) *Nature* 277:108 and Madzak (1992) *J Gen Virol* 73:1533; influenza virus, for example ATCC VR-797 and recombinant influenza viruses made employing reverse genetics techniques as described in US 5,166,057 and in Enami (1990) *Proc Natl Acad Sci* 87:3802-3805; Enami & Palese (1991) *J Virol* 65:2711-2713 and Luytjes (1989) *Cell* 59:110, (see also McMichael (1983) *NEJ Med* 309:13, and Yap (1978) *Nature* 273:238 and *Nature* (1979) 277:108); human immunodeficiency virus as described in EP-0386882 and in Buchschacher (1992) *J. Virol.* 66:2731; measles virus, for example ATCC VR-67 and VR-1247 and those described in EP-0440219; Aura virus, for example ATCC VR-368; Bebaru virus, for example ATCC VR-600 and ATCC VR-1240; Cabassou virus, for example ATCC VR-922; Chikungunya virus, for example ATCC VR-64 and ATCC VR-1241; Fort Morgan Virus, for example ATCC VR-924; Getah virus, for example ATCC VR-369 and ATCC VR-1243; Kyzylagach virus, for example ATCC VR-927; Mayaro virus, for example ATCC VR-66; Mucambo virus, for example ATCC VR-580 and ATCC VR-1244; Ndumu virus, for example ATCC VR-371; Pixuna virus, for example ATCC VR-372 and ATCC VR-1245; Tonate virus, for example

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ATCC VR-925; Trinit virus, for example ATCC VR-469; Una virus, for example ATCC VR-374; Whataroa virus, for example ATCC VR-926; Y-62-33 virus, for example ATCC VR-375; O'Nyong virus, Eastern encephalitis virus, for example ATCC VR-65 and ATCC VR-1242; Western encephalitis virus, for example ATCC VR-70, ATCC VR-1251, ATCC VR-622 and ATCC VR-1252; and coronavirus, for example ATCC VR-740 and those described in Hamre (1966) *Proc Soc Exp Biol Med* 121:190.

Delivery of the compositions of this invention into cells is not limited to the above mentioned viral vectors. Other delivery methods and media may be employed such as, for example, nucleic acid expression vectors, polycationic condensed DNA linked or unlinked to killed adenovirus alone, for example see US Serial No. 08/366,787, filed December 30, 1994 and Curiel (1992) *Hum Gene Ther* 3:147-154 ligand linked DNA, for example see Wu (1989) *J Biol Chem* 264:16985-16987, eucaryotic cell delivery vehicles cells, for example see US Serial No.08/240,030, filed May 9, 1994, and US Serial No. 08/404,796, deposition of photopolymerized hydrogel materials, hand-held gene transfer particle gun, as described in US Patent 5,149,655, ionizing radiation as described in US5,206,152 and in WO92/11033, nucleic charge neutralization or fusion with cell membranes. Additional approaches are described in Philip (1994) *Mol Cell Biol* 14:2411-2418 and in Woffendin (1994) *Proc Natl Acad Sci* 91:1581-1585.

Particle mediated gene transfer may be employed, for example see US Serial No. 60/023,867. Briefly, the sequence can be inserted into conventional vectors that contain conventional control sequences for high level expression, and then incubated with synthetic gene transfer molecules such as polymeric DNA-binding cations like polylysine, protamine, and albumin, linked to cell targeting ligands such as asialoorosomuroid, as described in Wu & Wu (1987) *J. Biol. Chem.* 262:4429-4432, insulin as described in Hucked (1990) *Biochem Pharmacol* 40:253-263, galactose as described in Plank (1992) *Bioconjugate Chem* 3:533-539, lactose or transferrin.

Naked DNA may also be employed to transform a host cell. Exemplary naked DNA introduction methods are described in WO 90/11092 and US 5,580,859. Uptake efficiency may be improved using biodegradable latex beads. DNA coated latex beads are efficiently transported into cells after endocytosis initiation by the beads. The method may be improved

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further by treatment of the beads to increase hydrophobicity and thereby facilitate disruption of the endosome and release of the DNA into the cytoplasm.

Liposomes that can act as gene delivery vehicles are described in U.S. 5,422,120, WO95/13796, WO94/23697, WO91/14445 and EP-524,968. As described in USSN. 60/023,867, on non-viral delivery, the nucleic acid sequences encoding a polypeptide can be inserted into conventional vectors that contain conventional control sequences for high level expression, and then be incubated with synthetic gene transfer molecules such as polymeric DNA-binding cations like polylysine, protamine, and albumin, linked to cell targeting ligands such as asialoorosomucoid, insulin, galactose, lactose, or transferrin. Other delivery systems include the use of liposomes to encapsulate DNA comprising the gene under the control of a variety of tissue-specific or ubiquitously-active promoters. Further non-viral delivery suitable for use includes mechanical delivery systems such as the approach described in Woffendin *et al* (1994) *Proc. Natl. Acad. Sci. USA* 91(24):11581-11585. Moreover, the coding sequence and the product of expression of such can be delivered through deposition of photopolymerized hydrogel materials. Other conventional methods for gene delivery that can be used for delivery of the coding sequence include, for example, use of hand-held gene transfer particle gun, as described in U.S. 5,149,655; use of ionizing radiation for activating transferred gene, as described in U.S. 5,206,152 and WO92/11033

Exemplary liposome and polycationic gene delivery vehicles are those described in US 5,422,120 and 4,762,915; in WO 95/13796; WO94/23697; and WO91/14445; in EP-0524968; and in Stryer, *Biochemistry*, pages 236-240 (1975) W.H. Freeman, San Francisco; Szoka (1980) *Biochem Biophys Acta* 600:1; Bayer (1979) *Biochem Biophys Acta* 550:464; Rivnay (1987) *Meth Enzymol* 149:119; Wang (1987) *Proc Natl Acad Sci* 84:7851; Plant (1989) *Anal Biochem* 176:420.

A polynucleotide composition can comprise a therapeutically effective amount of a gene therapy vehicle, as the term is defined above. For purposes of the present invention, an effective dose will be from about 0.01 mg/ kg to 50 mg/kg or 0.05 mg/kg to about 10 mg/kg of the DNA constructs in the individual to which it is administered.

Delivery Methods

Once formulated, the polynucleotide compositions of the invention can be administered (1) directly to the subject; (2) delivered *ex vivo*, to cells derived from the subject; or (3) *in vitro* for expression of recombinant proteins. The subjects to be treated can be mammals or birds. Also, human subjects can be treated.

Direct delivery of the compositions will generally be accomplished by injection, either subcutaneously, intraperitoneally, transdermally or transcutaneously, intravenously or intramuscularly or delivered to the interstitial space of a tissue. The compositions can also be administered into a tumor or lesion. Other modes of administration include oral and pulmonary administration, suppositories, and transdermal applications, needles, and gene guns or hyposprays. Dosage treatment may be a single dose schedule or a multiple dose schedule. See WO98/20734.

Methods for the *ex vivo* delivery and reimplantation of transformed cells into a subject are known in the art and described in e.g., WO93/14778. Examples of cells useful in *ex vivo* applications include, for example, stem cells, particularly hematopoietic, lymph cells, macrophages, dendritic cells, or tumor cells.

Generally, delivery of nucleic acids for both *ex vivo* and *in vitro* applications can be accomplished by the following procedures, for example, dextran-mediated transfection, calcium phosphate precipitation, polybrene mediated transfection, protoplast fusion, electroporation, encapsulation of the polynucleotide(s) in liposomes, and direct microinjection of the DNA into nuclei, all well known in the art.

Polynucleotide and Polypeptide pharmaceutical compositions

In addition to the pharmaceutically acceptable carriers and salts described above, the following additional agents can be used with polynucleotide and/or polypeptide compositions.

A. Polypeptides

One example are polypeptides which include, without limitation: asialoorosomucoid (ASOR); transferrin; asialoglycoproteins; antibodies; antibody fragments; ferritin; interleukins; interferons, granulocyte, macrophage colony stimulating factor (GM-CSF),

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granulocyte colony stimulating factor (G-CSF), macrophage colony stimulating factor (M-CSF), stem cell factor and erythropoietin. Viral antigens, such as envelope proteins, can also be used. Also, proteins from other invasive organisms, such as the 17 amino acid peptide from the circumsporozoite protein of plasmodium falciparum known as RII.

B. Hormones, Vitamins, Etc.

Other groups that can be included in a pharmaceutical composition include, for example: hormones, steroids, androgens, estrogens, thyroid hormone, or vitamins, folic acid.

C. Polyalkylenes, Polysaccharides, *etc.*

Also, polyalkylene glycol can be included in a pharmaceutical compositions with the desired polynucleotides and/or polypeptides. In a preferred embodiment, the polyalkylene glycol is polyethylene glycol. In addition, mono-, di-, or polysaccharides can be included. In a preferred embodiment of this aspect, the polysaccharide is dextran or DEAE-dextran. Also, chitosan and poly(lactide-co-glycolide) may be included in a pharmaceutical composition.

D. Lipids, and Liposomes

The desired polynucleotide or polypeptide can also be encapsulated in lipids or packaged in liposomes prior to delivery to the subject or to cells derived therefrom.

Lipid encapsulation is generally accomplished using liposomes which are able to stably bind or entrap and retain nucleic acid or polypeptide. The ratio of condensed polynucleotide to lipid preparation can vary but will generally be around 1:1 (mg DNA:micromoles lipid), or more of lipid. For a review of the use of liposomes as carriers for delivery of nucleic acids, see, Hug and Sleight (1991) *Biochim. Biophys. Acta.* 1097:1-17; Straubinger (1983) *Meth. Enzymol.* 101:512-527.

Liposomal preparations for use in the present invention include cationic (positively charged), anionic (negatively charged) and neutral preparations. Cationic liposomes have been shown to mediate intracellular delivery of plasmid DNA (Felgner (1987) *Proc. Natl. Acad. Sci. USA* 84:7413-7416); mRNA (Malone (1989) *Proc. Natl. Acad. Sci. USA* 86:6077-6081); and purified transcription factors (Debs (1990) *J. Biol. Chem.* 265:10189-10192), in functional form.

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Cationic liposomes are readily available. For example, N(1-2,3-dioleoyloxy)propyl)-N,N,N-triethylammonium (DOTMA) liposomes are available under the trademark Lipofectin, from GIBCO BRL, Grand Island, NY. (See, also, Felgner *supra*). Other commercially available liposomes include transfectace (DDAB/DOPE) and DOTAP/DOPE (Boehringer). Other cationic liposomes can be prepared from readily available materials using techniques well known in the art. See, e.g., Szoka (1978) *Proc. Natl. Acad. Sci. USA* 75:4194-4198; WO90/11092 for a description of the synthesis of DOTAP (1,2-bis(oleoyloxy)-3-(trimethylammonio)propane) liposomes.

Similarly, anionic and neutral liposomes are readily available, such as from Avanti Polar Lipids (Birmingham, AL), or can be easily prepared using readily available materials. Such materials include phosphatidyl choline, cholesterol, phosphatidyl ethanolamine, dioleoylphosphatidyl choline (DOPC), dioleoylphosphatidyl glycerol (DOPG), dioleoylphosphatidyl ethanolamine (DOPE), among others. These materials can also be mixed with the DOTMA and DOTAP starting materials in appropriate ratios. Methods for making liposomes using these materials are well known in the art.

The liposomes can comprise multilammellar vesicles (MLVs), small unilamellar vesicles (SUVs), or large unilamellar vesicles (LUVs). The various liposome-nucleic acid complexes are prepared using methods known in the art. See e.g., Straubinger (1983) *Meth. Immunol.* 101:512-527; Szoka (1978) *Proc. Natl. Acad. Sci. USA* 75:4194-4198; Papahadjopoulos (1975) *Biochim. Biophys. Acta* 394:483; Wilson (1979) *Cell* 17:77; Deamer & Bangham (1976) *Biochim. Biophys. Acta* 443:629; Ostro (1977) *Biochem. Biophys. Res. Commun.* 76:836; Fraley (1979) *Proc. Natl. Acad. Sci. USA* 76:3348; Enoch & Strittmatter (1979) *Proc. Natl. Acad. Sci. USA* 76:145; Fraley (1980) *J. Biol. Chem.* (1980) 255:10431; Szoka & Papahadjopoulos (1978) *Proc. Natl. Acad. Sci. USA* 75:145; and Schaefer-Ridder (1982) *Science* 215:166.

E. Lipoproteins

In addition, lipoproteins can be included with the polynucleotide or polypeptide to be delivered. Examples of lipoproteins to be utilized include: chylomicrons, HDL, IDL, LDL, and VLDL. Mutants, fragments, or fusions of these proteins can also be used. Also, modifications of naturally occurring lipoproteins can be used, such as acetylated LDL. These

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lipoproteins can target the delivery of polynucleotides to cells expressing lipoprotein receptors. Preferably, if lipoproteins are including with the polynucleotide to be delivered, no other targeting ligand is included in the composition.

Naturally occurring lipoproteins comprise a lipid and a protein portion. The protein portion are known as apoproteins. At the present, apoproteins A, B, C, D, and E have been isolated and identified. At least two of these contain several proteins, designated by Roman numerals, AI, AII, AIV; CI, CII, CIII.

A lipoprotein can comprise more than one apoprotein. For example, naturally occurring chylomicrons comprises of A, B, C, and E; over time these lipoproteins lose A and acquire C and E apoproteins. VLDL comprises A, B, C, and E apoproteins, LDL comprises apoprotein B; and HDL comprises apoproteins A, C, and E.

The amino acid sequences of these apoproteins are known and are described in, for example, Breslow (1985) *Annu Rev. Biochem* 54:699; Law (1986) *Adv. Exp Med. Biol.* 151:162; Chen (1986) *J Biol Chem* 261:12918; Kane (1980) *Proc Natl Acad Sci USA* 77:2465; and Utermann (1984) *Hum Genet* 65:232.

Lipoproteins contain a variety of lipids including, triglycerides, cholesterol (free and esters), and phospholipids. The composition of the lipids varies in naturally occurring lipoproteins. For example, chylomicrons comprise mainly triglycerides. A more detailed description of the lipid content of naturally occurring lipoproteins can be found, for example, in *Meth. Enzymol.* 128 (1986). The composition of the lipids are chosen to aid in conformation of the apoprotein for receptor binding activity. The composition of lipids can also be chosen to facilitate hydrophobic interaction and association with the polynucleotide binding molecule.

Naturally occurring lipoproteins can be isolated from serum by ultracentrifugation, for instance. Such methods are described in *Meth. Enzymol. (supra)*; Pitas (1980) *J. Biochem.* 255:5454-5460 and Mahey (1979) *J Clin. Invest* 64:743-750.

Lipoproteins can also be produced by *in vitro* or recombinant methods by expression of the apoprotein genes in a desired host cell. See, for example, Atkinson (1986) *Annu Rev Biophys Chem* 15:403 and Radding (1958) *Biochim Biophys Acta* 30: 443.

Lipoproteins can also be purchased from commercial suppliers, such as Biomedical Technologies, Inc., Stoughton, Massachusetts, USA.

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Further description of lipoproteins can be found in Zuckermann et al., PCT. Appln. No. US97/14465.

F. Polycationic Agents

Polycationic agents can be included, with or without lipoprotein, in a composition with the desired polynucleotide and/or polypeptide to be delivered.

Polycationic agents, typically, exhibit a net positive charge at physiological relevant pH and are capable of neutralizing the electrical charge of nucleic acids to facilitate delivery to a desired location. These agents have both in vitro, ex vivo, and in vivo applications. Polycationic agents can be used to deliver nucleic acids to a living subject either intramuscularly, subcutaneously, etc.

The following are examples of useful polypeptides as polycationic agents: polylysine, polyarginine, polyornithine, and protamine. Other examples of useful polypeptides include histones, protamines; human serum albumin, DNA binding proteins, non-histone chromosomal proteins, coat proteins from DNA viruses, such as Φ X174, transcriptional factors also contain domains that bind DNA and therefore may be useful as nucleic acid condensing agents. Briefly, transcriptional factors such as C/CEBP, c-jun, c-fos, AP-1, AP-2, AP-3, CPF, Prot-1, Sp-1, Oct-1, Oct-2, CREP, and TFIID contain basic domains that bind DNA sequences.

Organic polycationic agents include: spermine, spermidine, and putrescine.

The dimensions and of the physical properties of a polycationic agent can be extrapolated from the list above, to construct other polypeptide polycationic agents or to produce synthetic polycationic agents.

G. Synthetic Polycationic Agents

Synthetic polycationic agents which are useful in pharmaceutical compositions include, for example, DEAE-dextran, polybrene. Lipofectin™, and lipofectAMINE™ are monomers that form polycationic complexes when combined with polynucleotides or polypeptides.

Immunodiagnostic Assays

Neisseria MenB antigens, or antigenic fragments thereof, of the invention can be used in immunoassays to detect antibody levels (or, conversely, anti-*Neisseria* MenB antibodies can be used to detect antigen levels). Immunoassays based on well defined, recombinant antigens can be developed to replace invasive diagnostics methods. Antibodies to *Neisseria* MenB proteins or fragments thereof within biological samples, including for example, blood or serum samples, can be detected. Design of the immunoassays is subject to a great deal of variation, and a variety of these are known in the art. Protocols for the immunoassay may be based, for example, upon competition, or direct reaction, or sandwich type assays. Protocols may also, for example, use solid supports, or may be by immunoprecipitation. Most assays involve the use of labeled antibody or polypeptide; the labels may be, for example, fluorescent, chemiluminescent, radioactive, or dye molecules. Assays which amplify the signals from the probe are also known; examples of which are assays which utilize biotin and avidin, and enzyme-labeled and mediated immunoassays, such as ELISA assays.

Kits suitable for immunodiagnosis and containing the appropriate labeled reagents are constructed by packaging the appropriate materials, including the compositions of the invention, in suitable containers, along with the remaining reagents and materials (for example, suitable buffers, salt solutions, *etc.*) required for the conduct of the assay, as well as suitable set of assay instructions.

Nucleic Acid Hybridization

“Hybridization” refers to the association of two nucleic acid sequences to one another by hydrogen bonding. Typically, one sequence will be fixed to a solid support and the other will be free in solution. Then, the two sequences will be placed in contact with one another under conditions that favor hydrogen bonding. Factors that affect this bonding include: the type and volume of solvent; reaction temperature; time of hybridization; agitation; agents to block the non-specific attachment of the liquid phase sequence to the solid support (Denhardt's reagent or BLOTTO); concentration of the sequences; use of compounds to increase the rate of association of sequences (dextran sulfate or polyethylene glycol); and the

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stringency of the washing conditions following hybridization. See Sambrook *et al.* (*supra*) Volume 2, chapter 9, pages 9.47 to 9.57.

“Stringency” refers to conditions in a hybridization reaction that favor association of very similar sequences over sequences that differ. For example, the combination of temperature and salt concentration should be chosen that is approximately 120 to 200°C below the calculated T_m of the hybrid under study. The temperature and salt conditions can often be determined empirically in preliminary experiments in which samples of genomic DNA immobilized on filters are hybridized to the sequence of interest and then washed under conditions of different stringencies. See Sambrook *et al.* at page 9.50.

Variables to consider when performing, for example, a Southern blot are (1) the complexity of the DNA being blotted and (2) the homology between the probe and the sequences being detected. The total amount of the fragment(s) to be studied can vary a magnitude of 10, from 0.1 to 1 µg for a plasmid or phage digest to 10^{-9} to 10^{-8} g for a single copy gene in a highly complex eukaryotic genome. For lower complexity polynucleotides, substantially shorter blotting, hybridization, and exposure times, a smaller amount of starting polynucleotides, and lower specific activity of probes can be used. For example, a single-copy yeast gene can be detected with an exposure time of only 1 hour starting with 1 µg of yeast DNA, blotting for two hours, and hybridizing for 4-8 hours with a probe of 10^8 cpm/µg. For a single-copy mammalian gene a conservative approach would start with 10 µg of DNA, blot overnight, and hybridize overnight in the presence of 10% dextran sulfate using a probe of greater than 10^8 cpm/µg, resulting in an exposure time of ~24 hours.

Several factors can affect the melting temperature (T_m) of a DNA-DNA hybrid between the probe and the fragment of interest, and consequently, the appropriate conditions for hybridization and washing. In many cases the probe is not 100% homologous to the fragment. Other commonly encountered variables include the length and total G+C content of the hybridizing sequences and the ionic strength and formamide content of the hybridization buffer. The effects of all of these factors can be approximated by a single equation:

$$T_m = 81 + 16.6(\log_{10} C_i) + 0.4\%(G + C) - 0.6\%(\text{formamide}) - 600/n - 1.5\%(\text{mismatch})$$

where C_i is the salt concentration (monovalent ions) and n is the length of the hybrid in base pairs (slightly modified from Meinkoth & Wahl (1984) *Anal. Biochem.* 138:267-284).

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In designing a hybridization experiment, some factors affecting nucleic acid hybridization can be conveniently altered. The temperature of the hybridization and washes and the salt concentration during the washes are the simplest to adjust. As the temperature of the hybridization increases (i.e., stringency), it becomes less likely for hybridization to occur between strands that are nonhomologous, and as a result, background decreases. If the radiolabeled probe is not completely homologous with the immobilized fragment (as is frequently the case in gene family and interspecies hybridization experiments), the hybridization temperature must be reduced, and background will increase. The temperature of the washes affects the intensity of the hybridizing band and the degree of background in a similar manner. The stringency of the washes is also increased with decreasing salt concentrations.

In general, convenient hybridization temperatures in the presence of 50% formamide are 42°C for a probe with is 95% to 100% homologous to the target fragment, 37°C for 90% to 95% homology, and 32°C for 85% to 90% homology. For lower homologies, formamide content should be lowered and temperature adjusted accordingly, using the equation above. If the homology between the probe and the target fragment are not known, the simplest approach is to start with both hybridization and wash conditions which are nonstringent. If non-specific bands or high background are observed after autoradiography, the filter can be washed at high stringency and reexposed. If the time required for exposure makes this approach impractical, several hybridization and/or washing stringencies should be tested in parallel.

Nucleic Acid Probe Assays

Methods such as PCR, branched DNA probe assays, or blotting techniques utilizing nucleic acid probes according to the invention can determine the presence of cDNA or mRNA. A probe is said to "hybridize" with a sequence of the invention if it can form a duplex or double stranded complex, which is stable enough to be detected.

The nucleic acid probes will hybridize to the Neisserial nucleotide sequences of the invention (including both sense and antisense strands). Though many different nucleotide sequences will encode the amino acid sequence, the native Neisserial sequence is preferred because it is the actual sequence present in cells. mRNA represents a coding sequence and so

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a probe should be complementary to the coding sequence; single-stranded cDNA is complementary to mRNA, and so a cDNA probe should be complementary to the non-coding sequence.

The probe sequence need not be identical to the Neisserial sequence (or its complement) -- some variation in the sequence and length can lead to increased assay sensitivity if the nucleic acid probe can form a duplex with target nucleotides, which can be detected. Also, the nucleic acid probe can include additional nucleotides to stabilize the formed duplex. Additional Neisserial sequence may also be helpful as a label to detect the formed duplex. For example, a non-complementary nucleotide sequence may be attached to the 5' end of the probe, with the remainder of the probe sequence being complementary to a Neisserial sequence. Alternatively, non-complementary bases or longer sequences can be interspersed into the probe, provided that the probe sequence has sufficient complementarity with the a Neisserial sequence in order to hybridize therewith and thereby form a duplex which can be detected.

The exact length and sequence of the probe will depend on the hybridization conditions, such as temperature, salt condition and the like. For example, for diagnostic applications, depending on the complexity of the analyte sequence, the nucleic acid probe typically contains at least 10-20 nucleotides, preferably 15-25, and more preferably at least 30 nucleotides, although it may be shorter than this. Short primers generally require cooler temperatures to form sufficiently stable hybrid complexes with the template.

Probes may be produced by synthetic procedures, such as the triester method of Matteucci *et al.* (*J. Am. Chem. Soc.* (1981) 103:3185), or according to Urdea *et al.* (*Proc. Natl. Acad. Sci. USA* (1983) 80: 7461), or using commercially available automated oligonucleotide synthesizers.

The chemical nature of the probe can be selected according to preference. For certain applications, DNA or RNA are appropriate. For other applications, modifications may be incorporated e.g., backbone modifications, such as phosphorothioates or methylphosphonates, can be used to increase *in vivo* half-life, alter RNA affinity, increase nuclease resistance *etc.* (e.g., see Agrawal & Iyer (1995) *Curr Opin Biotechnol* 6:12-19; Agrawal (1996) *TIBTECH* 14:376-387); analogues such as peptide nucleic acids may also be

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used (e.g., see Corey (1997) *TIBTECH* 15:224-229; Buchardt *et al.* (1993) *TIBTECH* 11:384-386).

One example of a nucleotide hybridization assay is described by Urdea *et al.* in international patent application WO92/02526 (see also U.S. Patent 5,124,246).

Alternatively, the polymerase chain reaction (PCR) is another well-known means for detecting small amounts of target nucleic acids. The assay is described in: Mullis *et al.* (*Meth. Enzymol.* (1987) 155: 335-350); US patent 4,683,195; and US patent 4,683,202. Two "primer" nucleotides hybridize with the target nucleic acids and are used to prime the reaction. The primers can comprise sequence that does not hybridize to the sequence of the amplification target (or its complement) to aid with duplex stability or, for example, to incorporate a convenient restriction site. Typically, such sequence will flank the desired Neisserial sequence.

A thermostable polymerase creates copies of target nucleic acids from the primers using the original target nucleic acids as a template. After a threshold amount of target nucleic acids are generated by the polymerase, they can be detected by more traditional methods, such as Southern blots. When using the Southern blot method, the labeled probe will hybridize to the Neisserial sequence (or its complement).

Also, mRNA or cDNA can be detected by traditional blotting techniques described in Sambrook *et al* (*supra*). mRNA, or cDNA generated from mRNA using a polymerase enzyme, can be purified and separated using gel electrophoresis. The nucleic acids on the gel are then blotted onto a solid support, such as nitrocellulose. The solid support is exposed to a labeled probe and then washed to remove any unhybridized probe. Next, the duplexes containing the labeled probe are detected. Typically, the probe is labeled with a radioactive moiety.

EXAMPLES

The invention is based on the 961 nucleotide sequences from the genome of *N. meningitidis* set out in Appendix C, SEQ ID NOs:1-961 of the '573 application, which together represent substantially the complete genome of serotype B of *N. meningitidis*, as well as the full length genome sequence shown in Appendix D, SEQ ID NO 1068 of the '573

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application, and the full length genome sequence shown in Appendix A hereto, SEQ ID NO. 1.

It will be self-evident to the skilled person how this sequence information can be utilized according to the invention, as above described.

The standard techniques and procedures which may be employed in order to perform the invention (*e.g.* to utilize the disclosed sequences to predict polypeptides useful for vaccination or diagnostic purposes) were summarized above. This summary is not a limitation on the invention but, rather, gives examples that may be used, but are not required.

These sequences are derived from contigs shown in Appendix C (SEQ ID NOs 1-961) and from the full length genome sequence shown in Appendix D (SEQ ID NO 1068), which were prepared during the sequencing of the genome of *N. meningitidis* (strain B). The full length sequence was assembled using the TIGR Assembler as described by G.S. Sutton et al., *TIGR Assembler: A New Tool for Assembling Large Shotgun Sequencing Projects*, Genome Science and Technology, 1:9-19 (1995) [*see also* R. D. Fleischmann, et al., Science 269, 496-512 (1995); C. M. Fraser, et al., Science 270, 397-403 (1995); C. J. Bult, et al., Science 273, 1058-73 (1996); C. M. Fraser, et al., Nature 390, 580-586 (1997); J.-F. Tomb, et al., Nature 388, 539-547 (1997); H. P. Klenk, et al., Nature 390, 364-70 (1997); C. M. Fraser, et al., Science 281, 375-88 (1998); M. J. Gardner, et al., Science 282, 1126-1132 (1998); K. E. Nelson, et al., Nature 399, 323-9 (1999)]. Then, using the above-described methods, putative translation products of the sequences were determined. Computer analysis of the translation products were determined based on database comparisons. Corresponding gene and protein sequences, if any, were identified in *Neisseria meningitidis* (Strain A) and *Neisseria gonorrhoeae*. Then the proteins were expressed, purified, and characterized to assess their antigenicity and immunogenicity.

In particular, the following methods were used to express, purify, and biochemically characterize the proteins of the invention.

Chromosomal DNA Preparation

N. meningitidis strain 2996 was grown to exponential phase in 100 ml of GC medium, harvested by centrifugation, and resuspended in 5 ml buffer (20% Sucrose, 50 mM Tris-HCl, 50 mM EDTA, adjusted to pH 8.0). After 10 minutes incubation on ice, the bacteria were

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lysed by adding 10 ml lysis solution (50 mM NaCl, 1% Na-Sarkosyl, 50 µg/ml Proteinase K), and the suspension was incubated at 37°C for 2 hours. Two phenol extractions (equilibrated to pH 8) and one CHCl_3 /isoamylalcohol (24:1) extraction were performed. DNA was precipitated by addition of 0.3M sodium acetate and 2 volumes ethanol, and was collected by centrifugation. The pellet was washed once with 70% ethanol and redissolved in 4 ml buffer (10 mM Tris-HCl, 1mM EDTA, pH 8). The DNA concentration was measured by reading the OD at 260 nm.

Oligonucleotide design

Synthetic oligonucleotide primers were designed on the basis of the coding sequence of each ORF, using (a) the meningococcus B sequence when available, or (b) the gonococcus/meningococcus A sequence, adapted to the codon preference usage of meningococcus. Any predicted signal peptides were omitted, by deducing the 5'-end amplification primer sequence immediately downstream from the predicted leader sequence.

For most ORFs, the 5' primers included two restriction enzyme recognition sites (*Bam*HI-*Nde*I, *Bam*HI-*Nhe*I, or *Eco*RI-*Nhe*I, depending on the gene's restriction pattern); the 3' primers included a *Xho*I restriction site. This procedure was established in order to direct the cloning of each amplification product (corresponding to each ORF) into two different expression systems: pGEX-KG (using either *Bam*HI-*Xho*I or *Eco*RI-*Xho*I), and pET21b+ (using either *Nde*I-*Xho*I or *Nhe*I-*Xho*I).

5'-end primer tail:	<u>CGCGGATCCCATATG</u>	(<i>Bam</i> HI- <i>Nde</i> I)
	<u>CGCGGATCCGCTAGC</u>	(<i>Bam</i> HI- <i>Nhe</i> I)
	<u>CCGGAATTCTAGCTAGC</u>	(<i>Eco</i> RI- <i>Nhe</i> I)
3'-end primer tail:	<u>CCCGCTCGAG</u>	(<i>Xho</i> I)

For some ORFs, two different amplifications were performed to clone each ORF in the two expression systems. Two different 5' primers were used for each ORF; the same 3' *Xho*I primer was used as before:

5'-end primer tail:	<u>GGAATTCCATATGGCCATGG</u>	(<i>Nde</i> I)
5'-end primer tail:	<u>CGGGATCC</u>	(<i>Bam</i> HI)

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Other ORFs were cloned in the pTRC expression vector and expressed as an amino-terminus His-tag fusion. The predicted signal peptide may be included in the final product. *NheI*-*BamHI* restriction sites were incorporated using primers:

5'-end primer tail: GATCAGCTAGCCATATG (*NheI*)

3'-end primer tail: CGGGATCC (*BamHI*)

As well as containing the restriction enzyme recognition sequences, the primers included nucleotides which hybridized to the sequence to be amplified. The number of hybridizing nucleotides depended on the melting temperature of the whole primer, and was determined for each primer using the formulae:

$$T_m = 4 (G+C) + 2 (A+T) \quad (\text{tail excluded})$$

$$T_m = 64.9 + 0.41 (\% \text{ GC}) - 600/N \quad (\text{whole primer})$$

The average melting temperature of the selected oligos were 65-70°C for the whole oligo and 50-55°C for the hybridising region alone.

Oligos were synthesized by a Perkin Elmer 394 DNA/RNA Synthesizer, eluted from the columns in 2 ml $\text{NH}_4\text{-OH}$, and deprotected by 5 hours incubation at 56 °C. The oligos were precipitated by addition of 0.3M Na-Acetate and 2 volumes ethanol. The samples were then centrifuged and the pellets resuspended in either 100 μ l or 1ml of water. OD_{260} was determined using a Perkin Elmer Lambda Bio spectrophotometer and the concentration was determined and adjusted to 2-10 pmol/ μ l.

Table 1 shows the forward and reverse primers used for each amplification. In certain cases, it might be noted that the sequence of the primer does not exactly match the sequence in the ORF. When initial amplifications are performed, the complete 5' and/or 3' sequence may not be known for some meningococcal ORFs, although the corresponding sequences may have been identified in gonococcus. For amplification, the gonococcal sequences could thus be used as the basis for primer design, altered to take account of codon preference. In particular, the following codons may be changed: ATA \rightarrow ATT; TCG \rightarrow TCT; CAG \rightarrow CAA; AAG \rightarrow AAA; GAG \rightarrow GAA; CGA and CGG \rightarrow CGC; GGG \rightarrow GGC.

Amplification

The standard PCR protocol was as follows: 50-200 ng of genomic DNA were used as a template in the presence of 20-40 μ M of each oligo, 400-800 μ M dNTPs solution, 1x PCR

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buffer (including 1.5 mM MgCl₂), 2.5 units *TaqI* DNA polymerase (using Perkin-Elmer AmpliTaq, GIBCO Platinum, Pwo DNA polymerase, or Tahara Shuzo Taq polymerase).

In some cases, PCR was optimised by the addition of 10µl DMSO or 50 µl 2M betaine.

After a hot start (adding the polymerase during a preliminary 3 minute incubation of the whole mix at 95°C), each sample underwent a double-step amplification: the first 5 cycles were performed using as the hybridization temperature the one of the oligos excluding the restriction enzymes tail, followed by 30 cycles performed according to the hybridization temperature of the whole length oligos. The cycles were followed by a final 10 minute extension step at 72°C.

The standard cycles were as follows:

	Denaturation	Hybridisation	Elongation
First 5 cycles	30 seconds 95°C	30 seconds 50-55°C	30-60 seconds 72°C
Last 30 cycles	30 seconds 95°C	30 seconds 65-70°C	30-60 seconds 72°C

The elongation time varied according to the length of the ORF to be amplified.

The amplifications were performed using either a 9600 or a 2400 Perkin Elmer GeneAmp PCR System. To check the results, 1/10 of the amplification volume was loaded onto a 1-1.5% agarose gel and the size of each amplified fragment compared with a DNA molecular weight marker.

The amplified DNA was either loaded directly on a 1% agarose gel or first precipitated with ethanol and resuspended in a suitable volume to be loaded on a 1% agarose gel. The DNA fragment corresponding to the right size band was then eluted and purified from gel, using the Qiagen Gel Extraction Kit, following the instructions of the manufacturer. The final volume of the DNA fragment was 30µl or 50µl of either water or 10mM Tris, pH 8.5.

Digestion of PCR fragments

The purified DNA corresponding to the amplified fragment was split into 2 aliquots and double-digested with:

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NdeI/*Xho*I or *Nhe*I/*Xho*I for cloning into pET-21b+ and further expression of the protein as a C-terminus His-tag fusion

BamHI/*Xho*I or *Eco*RI/*Xho*I for cloning into pGEX-KG and further expression of the protein as a GST N-terminus fusion.

For ORF 76, *Nhe*I/*Bam*HI for cloning into pTRC-HisA vector and further expression of the protein as N-terminus His-tag fusion.

Each purified DNA fragment was incubated (37°C for 3 hours to overnight) with 20 units of each restriction enzyme (New England Biolabs) in a either 30 or 40 µl final volume in the presence of the appropriate buffer. The digestion product was then purified using the QIAquick PCR purification kit, following the manufacturer's instructions, and eluted in a final volume of 30 (or 50) µl of either water or 10mM Tris-HCl, pH 8.5. The final DNA concentration was determined by 1% agarose gel electrophoresis in the presence of titrated molecular weight marker.

Digestion of the cloning vectors (pET22B, pGEX-KG and pTRC-His A)

10 µg plasmid was double-digested with 50 units of each restriction enzyme in 200 µl reaction volume in the presence of appropriate buffer by overnight incubation at 37°C. After loading the whole digestion on a 1% agarose gel, the band corresponding to the digested vector was purified from the gel using the Qiagen QIAquick Gel Extraction Kit and the DNA was eluted in 50 µl of 10 mM Tris-HCl, pH 8.5. The DNA concentration was evaluated by measuring OD₂₆₀ of the sample, and adjusted to 50 µg/µl. 1 µl of plasmid was used for each cloning procedure.

Cloning

The fragments corresponding to each ORF, previously digested and purified, were ligated in both pET22b and pGEX-KG. In a final volume of 20 µl, a molar ratio of 3:1 fragment/vector was ligated using 0.5 µl of NEB T4 DNA ligase (400 units/µl), in the presence of the buffer supplied by the manufacturer. The reaction was incubated at room temperature for 3 hours. In some experiments, ligation was performed using the Boheringer "Rapid Ligation Kit", following the manufacturer's instructions.

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In order to introduce the recombinant plasmid in a suitable strain, 100 µl *E. coli* DH5 competent cells were incubated with the ligase reaction solution for 40 minutes on ice, then at 37°C for 3 minutes, then, after adding 800 µl LB broth, again at 37°C for 20 minutes. The cells were then centrifuged at maximum speed in an Eppendorf microfuge and resuspended in approximately 200 µl of the supernatant. The suspension was then plated on LB ampicillin (100 mg/ml).

The screening of the recombinant clones was performed by growing 5 randomly-chosen colonies overnight at 37 °C in either 2 ml (pGEX or pTC clones) or 5ml (pET clones) LB broth + 100 µg/ml ampicillin. The cells were then pelleted and the DNA extracted using the Qiagen QIAprep Spin Miniprep Kit, following the manufacturer's instructions, to a final volume of 30 µl. 5 µl of each individual miniprep (approximately 1g) were digested with either *NdeI/XhoI* or *BamHI/XhoI* and the whole digestion loaded onto a 1-1.5% agarose gel (depending on the expected insert size), in parallel with the molecular weight marker (1Kb DNA Ladder, GIBCO). The screening of the positive clones was made on the base of the correct insert size.

Cloning

Certain ORFs may be cloned into the pGEX-HIS vector using *EcoRI-PstI*, *EcoRI-SalI*, or *SalI-PstI* cloning sites. After cloning, the recombinant plasmids may be introduced in the *E.coli* host W3110.

Expression

Each ORF cloned into the expression vector may then be transformed into the strain suitable for expression of the recombinant protein product. 1 µl of each construct was used to transform 30 µl of *E.coli* BL21 (pGEX vector), *E.coli* TOP 10 (pTRC vector) or *E.coli* BL21-DE3 (pET vector), as described above. In the case of the pGEX-His vector, the same *E.coli* strain (W3110) was used for initial cloning and expression. Single recombinant colonies were inoculated into 2ml LB+Amp (100 µg/ml), incubated at 37°C overnight, then diluted 1:30 in 20 ml of LB+Amp (100 µg/ml) in 100 ml flasks, making sure that the OD₆₀₀ ranged between 0.1 and 0.15. The flasks were incubated at 30°C into gyratory water bath shakers until OD indicated exponential growth suitable for induction of expression (0.4-0.8 OD for

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pET and pTRC vectors; 0.8-1 OD for pGEX and pGEX-His vectors). For the pET, pTRC and pGEX-His vectors, the protein expression was induced by addition of 1mM IPTG, whereas in the case of pGEX system the final concentration of IPTG was 0.2 mM. After 3 hours incubation at 30°C, the final concentration of the sample was checked by OD. In order to check expression, 1ml of each sample was removed, centrifuged in a microfuge, the pellet resuspended in PBS, and analysed by 12% SDS-PAGE with Coomassie Blue staining. The whole sample was centrifuged at 6000g and the pellet resuspended in PBS for further use.

GST-fusion proteins large-scale purification.

A single colony was grown overnight at 37°C on LB+Amp agar plate. The bacteria were inoculated into 20 ml of LB+Amp liquid culture in a water bath shaker and grown overnight. Bacteria were diluted 1:30 into 600 ml of fresh medium and allowed to grow at the optimal temperature (20-37°C) to OD₅₅₀ 0.8-1. Protein expression was induced with 0.2mM IPTG followed by three hours incubation. The culture was centrifuged at 8000 rpm at 4°C. The supernatant was discarded and the bacterial pellet was resuspended in 7.5 ml cold PBS. The cells were disrupted by sonication on ice for 30 sec at 40W using a Branson sonifier B-15, frozen and thawed two times and centrifuged again. The supernatant was collected and mixed with 150µl Glutathione-Sepharose 4B resin (Pharmacia) (previously washed with PBS) and incubated at room temperature for 30 minutes. The sample was centrifuged at 700g for 5 minutes at 4°C. The resin was washed twice with 10 ml cold PBS for 10 minutes, resuspended in 1ml cold PBS, and loaded on a disposable column. The resin was washed twice with 2ml cold PBS until the flow-through reached OD₂₈₀ of 0.02-0.06. The GST-fusion protein was eluted by addition of 700µl cold Glutathione elution buffer (10mM reduced glutathione, 50mM Tris-HCl) and fractions collected until the OD₂₈₀ was 0.1. 21µl of each fraction were loaded on a 12% SDS gel using either Biorad SDS-PAGE Molecular weight standard broad range (M1) (200, 116.25, 97.4, 66.2, 45, 31, 21.5, 14.4, 6.5 kDa) or Amersham Rainbow Marker (M'') (220, 66, 46, 30, 21.5, 14.3 kDa) as standards. As the MW of GST is 26kDa, this value must be added to the MW of each GST-fusion protein.

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His-fusion soluble proteins large-scale purification.

A single colony was grown overnight at 37°C on a LB + Amp agar plate. The bacteria were inoculated into 20ml of LB+Amp liquid culture and incubated overnight in a water bath shaker. Bacteria were diluted 1:30 into 600ml fresh medium and allowed to grow at the optimal temperature (20-37°C) to OD₅₅₀ 0.6-0.8. Protein expression was induced by addition of 1 mM IPTG and the culture further incubated for three hours. The culture was centrifuged at 8000 rpm at 4°C, the supernatant was discarded and the bacterial pellet was resuspended in 7.5ml cold 10mM imidazole buffer (300 mM NaCl, 50 mM phosphate buffer, 10 mM imidazole, pH 8). The cells were disrupted by sonication on ice for 30 sec at 40W using a Branson sonifier B-15, frozen and thawed two times and centrifuged again. The supernatant was collected and mixed with 150µl Ni²⁺-resin (Pharmacia) (previously washed with 10mM imidazole buffer) and incubated at room temperature with gentle agitation for 30 minutes. The sample was centrifuged at 700g for 5 minutes at 4°C. The resin was washed twice with 10 ml cold 10mM imidazole buffer for 10 minutes, resuspended in 1ml cold 10mM imidazole buffer and loaded on a disposable column. The resin was washed at 4°C with 2ml cold 10mM imidazole buffer until the flow-through reached the O.D₂₈₀ of 0.02-0.06. The resin was washed with 2ml cold 20mM imidazole buffer (300 mM NaCl, 50 mM phosphate buffer, 20 mM imidazole, pH 8) until the flow-through reached the O.D₂₈₀ of 0.02-0.06. The His-fusion protein was eluted by addition of 700µl cold 250mM imidazole buffer (300 mM NaCl, 50 mM phosphate buffer, 250 mM imidazole, pH 8) and fractions collected until the O.D₂₈₀ was 0.1. 21µl of each fraction were loaded on a 12% SDS gel.

His-fusion insoluble proteins large-scale purification.

A single colony was grown overnight at 37 °C on a LB + Amp agar plate. The bacteria were inoculated into 20 ml of LB+Amp liquid culture in a water bath shaker and grown overnight. Bacteria were diluted 1:30 into 600ml fresh medium and let to grow at the optimal temperature (37°C) to O.D₅₅₀ 0.6-0.8. Protein expression was induced by addition of 1 mM IPTG and the culture further incubated for three hours. The culture was centrifuged at 8000rpm at 4°C. The supernatant was discarded and the bacterial pellet was resuspended in 7.5 ml buffer B (urea 8M, 10mM Tris-HCl, 100mM phosphate buffer, pH 8.8). The cells were disrupted by sonication on ice for 30 sec at 40W using a Branson sonifier B-15, frozen

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and thawed twice and centrifuged again. The supernatant was stored at -20°C, while the pellets were resuspended in 2 ml guanidine buffer (6M guanidine hydrochloride, 100mM phosphate buffer, 10 mM Tris-HCl, pH 7.5) and treated in a homogenizer for 10 cycles. The product was centrifuged at 13000 rpm for 40 minutes. The supernatant was mixed with 150µl Ni²⁺-resin (Pharmacia) (previously washed with buffer B) and incubated at room temperature with gentle agitation for 30 minutes. The sample was centrifuged at 700 g for 5 minutes at 4°C. The resin was washed twice with 10 ml buffer B for 10 minutes, resuspended in 1ml buffer B, and loaded on a disposable column. The resin was washed at room temperature with 2ml buffer B until the flow-through reached the OD₂₈₀ of 0.02-0.06. The resin was washed with 2ml buffer C (urea 8M, 10mM Tris-HCl, 100mM phosphate buffer, pH 6.3) until the flow-through reached the O.D₂₈₀ of 0.02-0.06. The His-fusion protein was eluted by addition of 700µl elution buffer (urea 8M, 10mM Tris-HCl, 100mM phosphate buffer, pH 4.5) and fractions collected until the OD₂₈₀ was 0.1. 21µl of each fraction were loaded on a 12% SDS gel.

His-fusion proteins renaturation

10% glycerol was added to the denatured proteins. The proteins were then diluted to 20µg/ml using dialysis buffer I (10% glycerol, 0.5M arginine, 50mM phosphate buffer, 5mM reduced glutathione, 0.5mM oxidised glutathione, 2M urea, pH 8.8) and dialysed against the same buffer at 4°C for 12-14 hours. The protein was further dialysed against dialysis buffer II (10% glycerol, 0.5M arginine, 50mM phosphate buffer, 5mM reduced glutathione, 0.5mM oxidised glutathione, pH 8.8) for 12-14 hours at 4°C. Protein concentration was evaluated using the formula:

$$\text{Protein (mg/ml)} = (1.55 \times \text{OD}_{280}) - (0.76 \times \text{OD}_{260})$$

Mice immunisations

20µg of each purified protein were used to immunise mice intraperitoneally. In the case of some ORFs, Balb-C mice were immunised with Al(OH)₃ as adjuvant on days 1, 21 and 42, and immune response was monitored in samples taken on day 56. For other ORFs, CD1 mice could be immunised using the same protocol. For other ORFs, CD1 mice could be immunised using Freund's adjuvant, and the same immunisation protocol was used, except that the immune response was measured on day 42, rather than 56. Similarly, for still other

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ORFs, CD1 mice could be immunised with Freund's adjuvant, but the immune response was measured on day 49.

ELISA assay (sera analysis)

The acapsulated MenB M7 strain was plated on chocolate agar plates and incubated overnight at 37°C. Bacterial colonies were collected from the agar plates using a sterile dracon swab and inoculated into 7ml of Mueller-Hinton Broth (Difco) containing 0.25% Glucose. Bacterial growth was monitored every 30 minutes by following OD₆₂₀. The bacteria were let to grow until the OD reached the value of 0.3-0.4. The culture was centrifuged for 10 minutes at 10000 rpm. The supernatant was discarded and bacteria were washed once with PBS, resuspended in PBS containing 0.025% formaldehyde, and incubated for 2 hours at room temperature and then overnight at 4°C with stirring. 100µl bacterial cells were added to each well of a 96 well Greiner plate and incubated overnight at 4°C. The wells were then washed three times with PBT washing buffer (0.1% Tween-20 in PBS). 200 µl of saturation buffer (2.7% Polyvinylpyrrolidone 10 in water) was added to each well and the plates incubated for 2 hours at 37°C. Wells were washed three times with PBT. 200 µl of diluted sera (Dilution buffer: 1% BSA, 0.1% Tween-20, 0.1% NaN₃ in PBS) were added to each well and the plates incubated for 90 minutes at 37°C. Wells were washed three times with PBT. 100 µl of HRP-conjugated rabbit anti-mouse (Dako) serum diluted 1:2000 in dilution buffer were added to each well and the plates were incubated for 90 minutes at 37°C. Wells were washed three times with PBT buffer. 100 µl of substrate buffer for HRP (25 ml of citrate buffer pH5, 10 mg of O-phenildiamine and 10 µl of H₂O) were added to each well and the plates were left at room temperature for 20 minutes. 100 µl H₂SO₄ was added to each well and OD₄₉₀ was followed. The ELISA was considered positive when OD490 was 2.5 times the respective pre-immune sera.

FACScan bacteria Binding Assay procedure.

The acapsulated MenB M7 strain was plated on chocolate agar plates and incubated overnight at 37°C. Bacterial colonies were collected from the agar plates using a sterile dracon swab and inoculated into 4 tubes containing 8ml each Mueller-Hinton Broth (Difco) containing 0.25% glucose. Bacterial growth was monitored every 30 minutes by following

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OD₆₂₀. The bacteria were let to grow until the OD reached the value of 0.35-0.5. The culture was centrifuged for 10 minutes at 4000 rpm. The supernatant was discarded and the pellet was resuspended in blocking buffer (1% BSA, 0.4% NaN₃) and centrifuged for 5 minutes at 4000 rpm. Cells were resuspended in blocking buffer to reach OD₆₂₀ of 0.07. 100µl bacterial cells were added to each well of a Costar 96 well plate. 100µl of diluted (1:200) sera (in blocking buffer) were added to each well and plates incubated for 2 hours at 4°C. Cells were centrifuged for 5 minutes at 4000 rpm, the supernatant aspirated and cells washed by addition of 200µl/well of blocking buffer in each well. 100µl of R-Phicoerytrin conjugated F(ab)₂ goat anti-mouse, diluted 1:100, was added to each well and plates incubated for 1 hour at 4°C. Cells were spun down by centrifugation at 4000rpm for 5 minutes and washed by addition of 200µl/well of blocking buffer. The supernatant was aspirated and cells resuspended in 200µl/well of PBS, 0.25% formaldehyde. Samples were transferred to FACScan tubes and read. The condition for FACScan setting were: FL1 on, FL2 and FL3 off; FSC-H Treshold:92; FSC PMT Voltage: E 02; SSC PMT: 474; Amp. Gains 7.1; FL-2 PMT: 539. Compensation values: 0.

OMV preparations

Bacteria were grown overnight on 5 GC plates, harvested with a loop and resuspended in 10 ml 20mM Tris-HCl. Heat inactivation was performed at 56°C for 30 minutes and the bacteria disrupted by sonication for 10' on ice (50% duty cycle, 50% output). Unbroken cells were removed by centrifugation at 5000g for 10 minutes and the total cell envelope fraction recovered by centrifugation at 50000g at 4°C for 75 minutes. To extract cytoplasmic membrane proteins from the crude outer membranes, the whole fraction was resuspended in 2% sarkosyl (Sigma) and incubated at room temperature for 20 minutes. The suspension was centrifuged at 10000g for 10 minutes to remove aggregates, and the supernatant further ultracentrifuged at 50000g for 75 minutes to pellet the outer membranes. The outer membranes were resuspended in 10mM Tris-HCl, pH8 and the protein concentration measured by the Bio-Rad Protein assay, using BSA as a standard.

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Whole Extracts preparation

Bacteria were grown overnight on a GC plate, harvested with a loop and resuspended in 1ml of 20mM Tris-HCl. Heat inactivation was performed at 56°C for 30' minutes.

Western blotting

Purified proteins (500ng/lane), outer membrane vesicles (5 µg) and total cell extracts (25µg) derived from MenB strain 2996 were loaded on 15% SDS-PAGE and transferred to a nitrocellulose membrane. The transfer was performed for 2 hours at 150mA at 4°C, in transferring buffer (0.3 % Tris base, 1.44 % glycine, 20% methanol). The membrane was saturated by overnight incubation at 4°C in saturation buffer (10% skimmed milk, 0.1% Triton X100 in PBS). The membrane was washed twice with washing buffer (3% skimmed milk, 0.1% Triton X100 in PBS) and incubated for 2 hours at 37°C with 1:200 mice sera diluted in washing buffer. The membrane was washed twice and incubated for 90 minutes with a 1:2000 dilution of horseradish peroxidase labeled anti-mouse Ig. The membrane was washed twice with 0.1% Triton X100 in PBS and developed with the Opti-4CN Substrate Kit (Bio-Rad). The reaction was stopped by adding water.

Bactericidal assay

MC58 strain was grown overnight at 37°C on chocolate agar plates. 5-7 colonies were collected and used to inoculate 7ml Mueller-Hinton broth. The suspension was incubated at 37°C on a nutator and let to grow until OD₆₂₀ was in between 0.5-0.8. The culture was aliquoted into sterile 1.5ml Eppendorf tubes and centrifuged for 20 minutes at maximum speed in a microfuge. The pellet was washed once in Gey's buffer (Gibco) and resuspended in the same buffer to an OD₆₂₀ of 0.5, diluted 1:20000 in Gey's buffer and stored at 25°C.

50µl of Gey's buffer/1% BSA was added to each well of a 96-well tissue culture plate. 25µl of diluted (1:100) mice sera (dilution buffer: Gey's buffer/0.2% BSA) were added to each well and the plate incubated at 4°C. 25µl of the previously described bacterial suspension were added to each well. 25µl of either heat-inactivated (56°C waterbath for 30 minutes) or normal baby rabbit complement were added to each well. Immediately after the addition of the baby rabbit complement, 22µl of each sample/well were plated on Mueller-

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Hinton agar plates (time 0). The 96-well plate was incubated for 1 hour at 37°C with rotation and then 22µl of each sample/well were plated on Mueller-Hinton agar plates (time 1). After overnight incubation the colonies corresponding to time 0 and time 1h were counted.

The following DNA and amino acid sequences are identified by titles of the following form: [g, m, or a] [#].[seq or pep], where "g" means a sequence from *N. gonorrhoeae*, "m" means a sequence from *N. meningitidis B*, and "a" means a sequence from *N. meningitidis A*; "#" means the number of the sequence; "seq" means a DNA sequence, and "pep" means an amino acid sequence. For example, "g001.seq" refers to an *N. gonorrhoeae* DNA sequence, number 1. The presence of the suffix "-1" or "-2" to these sequences indicates an additional sequence found for the same ORF. Further, open reading frames are identified as ORF #, where "#" means the number of the ORF, corresponding to the number of the sequence which encodes the ORF, and the ORF designations may be suffixed with ".ng" or ".a", indicating that the ORF corresponds to a *N. gonorrhoeae* sequence or a *N. meningitidis A* sequence, respectively. Computer analysis was performed for the comparisons that follow between "g", "m", and "a" peptide sequences; and therein the "pep" suffix is implied where not expressly stated.

EXAMPLE 1

The following ORFs were predicted from the contig sequences and/or the full length sequences using the methods herein described.

Localization of the ORFs

ORF: contig:

279 gnm4.seq

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 2>:

m279.seq

```

1   ATAACGCGGA TTTGCGGCTG CTTGATTTC ACGGTTTTC GGGCTTCGGC
51  AAGTTTGTCT GCGGCGGGTT TCATCAGGCT GCAATGGGAA GGTACGGACA
101 CGGGCAGCGG CAGGGCGCGT TTGGCACCGG CTTCTTTGGC GGCAGCCATG
151 GCGCGTCCGA CGGCGGCGGC GTTGCTTGCA ATCACGATTT GTCCGGGTGA
201 GTTGAAGTTG ACGGCTTCGA CCACTTCGCT TTGGGCGGCT TCGGCACAAA
251 TGGCTTTAAC CTGCTCATCT TCCAAGCCGA GAATCGCCGC CATTGCGCCC
301 ACGCCTTGCG GTACGGCGGA CTGCATCAGT TCGGCGCGCA GGCGCACGAG
351 TTTGACCGCG TCGGCAAAAT TCAATGCGCC GCGGCAACG AGTGCGGTGT
401 ATTCGCCGAG GCTGTGTCCG GCAACGGCGG CAGGCGTTTT GCCGCCGCT
451 TCTAAATAG
```

This corresponds to the amino acid sequence <SEQ ID 3; ORF 279>:

m279.pep

```

1  ITRICGLIS TVFRASASLS AAGFIRLQWE GTDTGSGRAR LAPASLAAAM
51  ARPTAAALPA ITICPGELKL TASTTSLWAA SAQMALTCSS SKPRIAAIAP
101 TPCGTADCIS SARRRTSLTA SAKFNAPAAAT SAVYSPRLCP ATAAGVLPPA
151 SK*

```

The following partial DNA sequence was identified in *N.gonorrhoeae* <SEQ ID 4>:

g279.req

1	atgacgcgga	tttgcggtg	cttgatttca	acggttttga	gtgtttcggc
51	aagtttgtcg	gcggcgggtt	tcacagggt	gcaatgggaa	ggaacggata
101	ccggcagcgg	cagggcgcg	ttggctccgg	cttctttggc	ggcagccatg
151	gtgcttccga	cggcggcggc	gttgcttgca	atcacgactt	gtccgggcga
201	gttgaagtgt	acggcttcga	ccacttcgcc	ctgtgcggat	tcggcacaaa
251	tctgcctgac	ctgttcatct	tccaaaccca	aaatggccgc	cattgcgctt
301	acgccttgcg	gtacggcgga	ctgcatcagt	tcggcgcgca	ggcggacgag
351	tttgacggca	tcggcaaaa	ccaatgcttc	ggcggcgaca	agcgcggtgt
401	attcgccgag	gctgtgtccg	gcaacggcgg	caggcgtttt	gccgccact
451	tccaaataq				

This corresponds to the amino acid sequence <SEQ ID 5; ORF 279.ng>:

g279.pep

```

1  MTRICGCLIS TVLSVSASLS AAGFIRLQWE GTDTGSGRAR LAPASLAAAM
51  VRPTAAALPA ITTCPGELKL TASTTSPCAD SAQICLTCSS SKPKMAAIAP
101 TPCGTADCIS SARRRTSLTA SAKSNASAAT SAVYSPRLCP ATAAGVLPPT
151 SK*

```

ORF 279 shows 89.5% identity over a 152 aa overlap with a predicted ORF (ORF 279.ng) from *N. gonorrhoeae*:

```

m279.pep      10      20      30      40      50      60
               ITRICGCLISTVFRASASLSAAGFIRLQWEGTDTGSGRARLAPASLAAAMARPTAAALPA
               :|||||: :|||:|||||:|||||:|||||:|||||:|||||:|||||:|||||:|||||:
g279           MTRICGCLISTVLSVSASLSAAGFIRLQWEGTDTGSGRARLAPASLAAAMVRPTAAALPA
               10      20      30      40      50      60

m279.pep      70      80      90      100     110     120
               ITICPGELKLTASTTSLWAASAQMALTCSSSKPRIAAIAPTTCGTADCISSARRRTSLTA
               |||:|||||:|||||:|||||:|||||:|||||:|||||:|||||:|||||:|||||:
g279           ITTCPGELKLTASTTSPCADSAQICLTCSSSKPKMAAIAPTTCGTADCISSARRRTSLTA
               70      80      90      100     110     120

m279.pep      130     140     150
               SAKFNAPAATSAVYSPRLCPATAAGVLPPASKX
               |||:|||||:|||||:|||||:|||||:|||||:|||||:|||||:|||||:
g279           SAKSNASAATSAVYSPRLCPATAAGVLPPTSKX
               130     140     150

```

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 6>:

a279.seq

1	ATGACNCNGA	TTTGGCGGCTG	CTTGATTTC	ACGGTTTNNA	GGGCTTCGGC
51	GAGTTTGTCG	GCGCGGGGTT	TCATGAGGCT	GCAATTGGGA	GGTACNGACA
101	CNGCGACGCG	CAGGGCGCGT	TGTGCGCCGG	CTTCTTTGGC	GGCAAGCATA
151	CGCGCGCTCGA	CGCGCGCGGC	ATTGCTCTGCA	ATCACGACTT	TCGCGGCGGA
201	GTTGAAGTTG	ACGGCTTCAA	CCACTTCATC	CTGTGCGGAT	TCGGCGCAAA
251	TTTGCTTTTAC	CTGTTCATCT	TCCAAGCCGA	GAATCGCCGC	CATTGCGCCC
301	ACGCGTTTGGC	TCAGGCGCGA	CTGCATCAGT	TGGCGCGCGC	NGTCGACGAG
351	TTTGACCGCG	TGCGCAAAAT	CCAATGCGCC	GGCGGCAACN	AGTGCGGTGT

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401 ATTCGCCGAN GCTGTGTCCG GCAACGGCGG CAGGCGTTTT GCCGCCCGCT
451 TCCGAATAG

This corresponds to the amino acid sequence <SEQ ID 7; ORF 279.a>:

a279.pep
1 MTXICGCLIS TVXRASASLS AAGFMRLQWE GTDTGSGRAR LAPASLAASI
51 ARSTAAALPA ITTCPGELKL TASTSSCAD SAQICFTCSS SKPRIAIAIP
101 TPCGTADCIS SARXRTSLTA SAKSNAPAAT SAVYSPXLCP ATAAGVLPPA
151 SE*

m279/a279 ORFs 279 and 279.a showed a 88.2% identity in 152 aa overlap

	10	20	30	40	50	60
m279.pep	ITRICGCLISTVFRASASLSAAGFIRLQWEGTDTGSGRARLAPASLAAAMARPTAAALPA					
	: : :					
a279	MTXICGCLISTVXRASASLSAAGFMRLQWEGTDTGSGRARLAPASLAASIARSTAAALPA					
	10	20	30	40	50	60
	70	80	90	100	110	120
m279.pep	ITICPGELKLTASTTSLWAASAQMALTCSSSKPRIAAIAPTPCGTADCISSARRRTSLTA					
	:					
a279	ITTCPGELKLTASTTSSCADSAQICFTCSSSKPRIAAIAPTPCGTADCISSARXRTSLTA					
	70	80	90	100	110	120
	130	140	150			
m279.pep	SAKFNAPAATSAVYSPRLCPATAAGVLPPASKX					
	:					
a279	SAKSNAPAATSAVYSPXLCPATAAGVLPPASEX					
	130	140	150			

519 and 519-1 gnm7.seq

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 8>:

m519.seq (partial)
1 ..TCCGTTATCG GCGTATGGA GTTGGACAAA ACGTTTGAAG AACGCGACGA
51 AATCAACAGT ACTGTTGTTG CGGCTTTGGA CGAGGCGGCC GGGgCTTgGG
101 GTGTGAAGGT TTTGCGTTAT GAGATTAAAG ACTTGTTCC GCCGCAAGAA
151 ATCCTTCGCT CAATGCAGGC GCAAATTACT GCCGAACGCG AAAAACGCGC
201 CCGTATCGCC GAATCCGAAG GTCGTAAAT CGAACAATC AACCTTGCCA
251 GTGGTCAGCG CGAAGCCGAA ATCCAACAAT CCGAAGGCGA GGCTCAGGCT
301 GCGGTCAATG CGTCAAATGC CGAGAAAATC GCCCGCATCA ACCGCGCCAA
351 AGGTGAAGCG GAATCCTTGC GCCTTGTTGC CGAAGCCAAT GCCGAAGCCA
401 TCCGTCAAAT TGCCGCCGCC CTTCAAACCC AAGGCGGTGC GGATGCGGTC
451 AATCTGAAGA TTGCGGAACA ATACGTCGCT GCGTTCAACA ATCTTGCCAA
501 AGAAAGCAAT ACGCTGATTA TGCCCGCCAA TGTTGCCGAC ATCGGCAGCC
551 TGATTTCTGC CGGTATGAAA ATTATCGACA GCAGCAAAAC CGCCAAaTAA

This corresponds to the amino acid sequence <SEQ ID 9; ORF 519>:

m519.pep (partial)
1 ..SVIGRMELDK TFEERDEINS TVVAALDEAA GAWGVKVLRY EIKDLVPPQE
51 ILRSMQAQIT AEREKRARIA ESEGRKIEQI NLASGQREAE IQQSEGEAQA
101 AVNASNAEKI ARINRAKGEA ESLRLVAEAN AEAIROIAAA LQTQGGADAV
151 NLKIAEQYVA AFNNLAKESN TLIMPANVAD IGSLISAGMK IIDSSKTAK*

The following partial DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 10>:

g519.seq
1 atggaatttt tcattatctt gttggcagcc gtcgcccgttt tcggcttcaa
51 atcctttgtc gtcaccccc agcaggaagt ccacgttgtc gaaaggctcg

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```

101  ggcgtttcca  tcgcgccctg  acggccgggt  tgaatatttt  gattcccttt
151  atcgaccgcg  tcgcctaccg  ccattcgctg  aaagaaatcc  ctttagacgt
201  acccagccag  gtctgcatca  cgcgcgataa  tacgcaattg  actgttgacg
251  gcatcatcta  tttccaagta  accgatccca  aactcgcttc  atacggttcg
301  agcaactaca  ttatggcaat  taccagcgtt  gcccaaacga  cgctgcgttc
351  cgttatcggg  cgtatggagt  tggacaaaac  gtttgaagaa  cgcgacgaaa
401  tcaacagtac  cgtcgtctcc  gccctcgatg  aagccgcggg  ggcttggggg
451  gtgaaagtcc  tccgttacga  aatcaaggat  ttggttccgc  cgcaagaaat
501  ctttcgcgca  atgcaggcac  aaattaccgc  cgaacgcgaa  aaacgcgccc
551  gtattgccga  atccgaaggc  cgtaaaatcg  aacaaatcaa  ctttgccagt
601  ggtcagcgtg  aagccgaaat  ccaacaatcc  gaaggcgagg  ctcaaggctgc
651  ggtcaatgcg  tccaatgccg  agaaaatcgc  ccgcatcaac  cgcgccaaag
701  gcgaagcgga  atccctgcgc  cttgttgccg  aagccaatgc  cgaagccaac
751  cgtcaaattg  ccgcccgcct  tcaaacccaa  agcggggcgg  atgcggtcaa
801  tctgaagatt  gcgggacaat  acgttaccgc  gttcaaaaat  cttgccaaaag
851  aagacaatac  gcggattaag  cccgccaaag  ttgccgaaat  cgggaaccct
901  aattttcggc  ggcatgaaaa  attttcgcca  gaagcaaaaa  cggccaaata
951  a

```

This corresponds to the amino acid sequence <SEQ ID 11; ORF 519.ng>:

```

g519.pep
  1  MEFFIILLAA  VAVFGFKSFV  VIPQQEVHVV  ERLGRFHRAL  TAGLNILIPF
 51  IDRWAYRHSL  KEIPLDVPSQ  VCITRDNTQL  TVDGIYFQV  TDPKLASYGS
101  SNYIMAITQL  AQTTLRSVIG  RMELDKTFEE  RDEINSTVVS  ALDEAAGAWG
151  VKVLRYEIKD  LVPPQEILRA  MQAQITAERE  KRARIAESEG  RKIEQINLAS
201  GQREAEIQQS  EGEEAQAANA  SNAEKIARIN  RAKGEAESLR  LVAEANAEAN
251  RQIAAALQTQ  SGADAVNLKI  AGQYVTAFKN  LAKEDNTRIK  PAKVAEIGNP
301  NFRRHEKFSP  EAKTAK*

```

ORF 519 shows 87.5% identity over a 200 aa overlap with a predicted ORF (ORF 519.ng) from *N. gonorrhoeae*:

```

m519/g519

m519.pep                                10      20      30
                                SVIGRMELDKTFEERDEINSTVVAALDEAA
                                |||
g519      YFQVTDPKLASYGSSNYIMAITQLAQTTLRSVIGRMELDKTFEERDEINSTVVSALDEAA
              90      100      110      120      130      140

m519.pep                                40      50      60      70      80      90
                                GAWGVKVLRYEIKDLVPPQEILRSMQAITAEREKRARIAESEGRKIEQINLASGQREAE
                                |||
g519      GAWGVKVLRYEIKDLVPPQEILRAMQAITAEREKRARIAESEGRKIEQINLASGQREAE
              150      160      170      180      190      200

m519.pep                                100      110      120      130      140      150
                                IQQSEGEAQAANASNAEKIARINRAKGEAESLRLVAEANAEAIRQIAAALQTQGGADAV
                                |||
g519      IQQSEGEAQAANASNAEKIARINRAKGEAESLRLVAEANAEANRQIAAALQTQSGADAV
              210      220      230      240      250      260

m519.pep                                160      170      180      190      200
                                NLKIAEQYVAAFNNLAKESNTLIMPANVADIGSL- ISAGMKIIDSSKTAK
                                |||
g519      NLKIAEQYVTAFKNLAKEDNTRIKPAKVAEIGNPNFRRHEKFSPEAKTAK
              270      280      290      300      310

```

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 12>:

a519.seq

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```

1  ATGGAATTTT TCATTATCTT GCTGGCAGCC GTCGTTGTTT TCGGCTTCAA
51  ATCCTTTGTT GTCATCCAC  AGCAGGAAGT CCACGTTGTC GAAAGGCTCG
101 GCGGTTTCCA TCGCGCCCTG ACGGCCGGTT TGAATATTTT GATTCCTTTT
151 ATCGACCGCG TCGCCTACCG CCATTCGCTG AAAGAAATCC CTTTAGACGT
201 ACCCAGCCAG GTCTGCATCA CGCGCGACAA TACGCAGCTG ACTGTTGACG
251 GTATCATCTA TTTCCAAGTA ACCGACCCCA AACTCGCCTC ATACGGTTCG
301 AGCAACTACA TTATGGCGAT TACCCAGCTT GCCCAAACGA CGCTGCGTTC
351 CGTTATCGGG CGTATGGAAT TGGACAAAAC GTTTGAAGAA CGCGACGAAA
401 TCAACAGCAC CGTCGTCTCC GCCCTCGATG AAGCCGCCGG AGCTTGGGGT
451 GTGAAGGTTT TGCGTTATGA GATTAAAGAC TTGGTTCGCG CGCAAGAAAT
501 CCTTCGCTCA ATGCAGGCGC AAATTACTGC TGAACGCGAA AAACGCGCCC
551 GTATCGCCGA ATCCGAAGGT CGTAAAATCG AACAAATCAA CCTTGCCAGT
601 GGTCAGCGCG AAGCCGAAAT CCAACAATCC GAAGGCGAGG CTCAGGTGCG
651 GGTCAATGCG TCAAATGCCG AGAAAATCGC CCGCATCAAC CGCGCCAAAG
701 GTGAAGCGGA ATCCTTGCGC CTTGTTGCCG AAGCCAATGC CGAAGCCATC
751 CGTCAAATG CCGCCGCCCT TCAAACCCAA GGCGGTGCGG ATGCGGTCAA
801 TCTGAAGATT GCGGAACAAT ACGTCGCCGC GTTCAACAAT CTTGCCAAAG
851 AAAGCAATAC GCTGATTATG CCGCCAATG TTGCCGACAT CGGCAGCCTG
901 ATTTCTGCCG GTATGAAAT TATCGACAGC AGCAAAACCG CCAAATAA

```

This corresponds to the amino acid sequence <SEQ ID 13; ORF 519.a>:

```

a519.pep
1  MEFFIILLAA VVVFQFKSFV VIPQQEVHVV ERLGRFHRAL TAGLNILIPF
51  IDRVAIRHSL KEIPLDVPSQ VCITRDNTQL TVDGLIYFQV TDPKLASYGS
101 SNYIMAITQL AQTTLRSVIG RMELDKTFEE RDEINSTVVS ALDEAAGAWG
151 VKVLRYEIKD LVPPQEILRS MQAQITAERE KRARIAESEG RKIEQINLAS
201 GQREAEIQQS EGEAQAAVNA SNAEKIARIN RAKGEAESLR LVAEANAEAI
251 RQIAAALQTQ GGADAVNLKI AEQYVAAFNN LAKESNTLIM PANVADIGSL
301 ISAGMKIIDS SKTAK*

```

m519/a519 ORFs 519 and 519.a showed a 99.5% identity in 199 aa overlap

```

                                     10      20      30
m519.pep                               SVIGRMELDKTFEERDEINSTVVAALDEAA
a519      YFQVTDPKLASYGSSNYIMAITQLAQTTLRSVIGRMELDKTFEERDEINSTVVSALDEAA
           90      100      110      120      130      140

                                     40      50      60      70      80      90
m519.pep      GAWGVKVLRYEIKDLVPPQEILRSMQQAQITAEREKRARIAESEGRKIEQINLASGQREAE
a519      GAWGVKVLRYEIKDLVPPQEILRSMQQAQITAEREKRARIAESEGRKIEQINLASGQREAE
           150      160      170      180      190      200

                                     100      110      120      130      140      150
m519.pep      IQQSEGEAQAAVNASNAEKIARINRAKGEAESLRLVAEANAEAIRQIAAALQTQGGADAV
a519      IQQSEGEAQAAVNASNAEKIARINRAKGEAESLRLVAEANAEAIRQIAAALQTQGGADAV
           210      220      230      240      250      260

                                     160      170      180      190      200
m519.pep      NLKIAEQYVAAFNNLAKESNTLIMPANVADIGSLISAGMKIIDSSKTAKX
a519      NLKIAEQYVAAFNNLAKESNTLIMPANVADIGSLISAGMKIIDSSKTAKX
           270      280      290      300      310

```

Further work revealed the following DNA sequence identified in *N. meningitidis* <SEQ ID 14>:

m519-1.seq

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```

1  ATGGAATTTT TCATTATCTT GTTGGTAGCC GTCGCCGTTT TCGGTTTCAA
51  ATCCTTTGTT GTCATCCAC AACAGGAAGT CCACGTTGTC GAAAGGCTGG
101 GCGTTTTCCA TCGCGCCCTG ACGGCGGTT TGAATATTTT GATTCCCTTT
151 ATCGACCGCG TCGCCTACCG CCATTCGCTG AAAGAAATCC CTTTAGACGT
201 ACCCAGCCAG GTCTGCATCA CGCGCGACAA TACGCAGCTG ACTGTTGACG
251 GCATCATCTA TTTCCAAGTA ACCGACCCCA AACTCGCCTC ATACGGTTCG
301 AGCAACTACA TTATGGCGAT TACCCAGCTT GCCCAAACGA CGCTGCGTTC
351 CGTTATCGGG CGTATGGAGT TGGACAAAAC GTTTGAAGAA CGCGACGAAA
401 TCAACAGTAC TGTGTGTGCG GCTTGGACG AGGCGGCCGG GGCTTGGGGT
451 GTGAAGGTTT TGCCTTATGA GATTAAAGAC TTGGTTCCGC CGCAAGAAAT
501 CCTTCGCTCA ATGCAGGCGC AAATTACTGC CGAACGCGAA AAACGCGCCC
551 GTATCGCCGA ATCCGAAGGT CGTAAATCG AACAAATCAA CCTTGCCAGT
601 GGTCAGCGCG AAGCCGAAAT CCAACAATCC GAAGGCGAGG CTCAGGCTGC
651 GGTCAATGCG TCAATGCCG AGAAAATCGC CCGCATCAAC CGCGCCAAAG
701 GTGAAGCGGA ATCCTTGCGC CTTGTTGCCG AAGCCAATGC CGAAGCCATC
751 CGTCAAATTG CCGCCGCCCT TCAAACCCAA GGCGGTGCGG ATGCGGTCAA
801 TCTGAAGATT GCGGAACAAT ACGTCGCTGC GTTCAACAAT CTTGCCAAAG
851 AAAGCAATAC GCTGATTATG CCCGCCAATG TTGCCGACAT CGGCAGCCTG
901 ATTTCTGCCG GTATGAAAT TATCGACAGC AGCAAAACCG CCAAATAA

```

This corresponds to the amino acid sequence <SEQ ID 15; ORF 519-1>:

m519-1.

```

1  MEFFIILLVA VAVFGFKSFV VIPQQEVHVV ERLGRFHRAL TAGLNILIPF
51  IDRVAYRHSI KEIPLDVPSQ VCI TRDNTQL TVDGIIFYQV TDPKLSYGS
101 SNYIMAITQL AQTTLRSVIG RMELDKTFEE RDEINSTVVA ALDEAAGAWG
151 VKVLRYEIKD LVPPQEILRS MQAQITAERE KRARIAESEG RKIEQINLAS
201 GQREAEIQQS EGEAQA AVNA SNAEKIARIN RAKGEAESLR LVAEANA EAI
251 RQIAAALQTQ GGADAVNLKI AEQYVAAFNN LAKESNTLIM PANVADIGSL
301 ISAGMKIIDS SKTAK*

```

The following DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 16>:

g519-1.seq

```

1  ATGGAATTTT TCATTATCTT GTTGGCAGCC GTCGCCGTTT TCGGCTTCAA
51  ATCCTTTGTC GTCATCCCCC AGCAGGAAGT CCACGTTGTC GAAAGGCTCG
101 GCGTTTTCCA TCGCGCCCTG ACGGCGGTT TGAATATTTT GATTCCCTTT
151 ATCGACCGCG TCGCCTACCG CCATTCGCTG AAAGAAATCC CTTTAGACGT
201 ACCCAGCCAG GTCTGCATCA CGCGCGATAA TACGCAATTG ACTGTTGACG
251 GCATCATCTA TTTCCAAGTA ACCGATCCCA AACTCGCCTC ATACGGTTCG
301 AGCAACTACA TTATGGCAAT TACCCAGCTT GCCCAAACGA CGCTGCGTTC
351 CGTTATCGGG CGTATGGAGT TGGACAAAAC GTTTGAAGAA CGCGACGAAA
401 TCAACAGTAC CGTCGTCTCC GCCCTCGATG AAGCCGCCGG GGCTTGGGGT
451 GTGAAAGTCC TCCGTTACGA AATCAAGGAT TTGGTTCCGC CGCAAGAAAT
501 CCTTCGCGCA ATGCAGGCAC AAATTACCGC CGAACGCGAA AAACGCGCCC
551 GTATTGCCGA ATCCGAAGGC CGTAAATCG AACAAATCAA CCTTGCCAGT
601 GGTCAGCGTG AAGCCGAAAT CCAACAATCC GAAGGCGAGG CTCAGGCTGC
651 GGTCAATGCG TCCAATGCCG AGAAAATCGC CCGCATCAAC CGCGCCAAAG
701 GCGAAGCGGA ATCCCTGCGC CTTGTTGCCG AAGCCAATGC CGAAGCCATC
751 CGTCAAATTG CCGCCGCCCT TCAAACCCAA GGCGGGGCGG ATGCGGTCAA
801 TCTGAAGATT GCGGAACAAT ACGTAGCCGC GTTCAACAAT CTTGCCAAAG
851 AAAGCAATAC GCTGATTATG CCCGCCAATG TTGCCGACAT CGGCAGCCTG
901 ATTTCTGCCG GCATGAAAT TATCGACAGC AGCAAAACCG CCAAATAA

```

This corresponds to the amino acid sequence <SEQ ID 17; ORF 519-1.ng>:

g519-1.pep

```

1  MEFFIILLAA VAVFGFKSFV VIPQQEVHVV ERLGRFHRAL TAGLNILIPF
51  IDRVAYRHSI KEIPLDVPSQ VCI TRDNTQL TVDGIIFYQV TDPKLSYGS
101 SNYIMAITQL AQTTLRSVIG RMELDKTFEE RDEINSTVVS ALDEAAGAWG
151 VKVLRYEIKD LVPPQEILRA MQAQITAERE KRARIAESEG RKIEQINLAS
201 GQREAEIQQS EGEAQA AVNA SNAEKIARIN RAKGEAESLR LVAEANA EAI
251 RQIAAALQTQ GGADAVNLKI AEQYVAAFNN LAKESNTLIM PANVADIGSL
301 ISAGMKIIDS SKTAK*

```

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m519-1/g519-1 ORFs 519-1 and 519-1.ng showed a 99.0% identity in 315 aa overlap

	10	20	30	40	50	60
g519-1.pep	MEFFIILLAAVAVFGFKSFVVI PQQEVHVVERLGRFHRALTAGLNILIPFIDRVAYRHSL					
	:					
m519-1	MEFFIILLVAVAVFGFKSFVVI PQQEVHVVERLGRFHRALTAGLNILIPFIDRVAYRHSL					
	10	20	30	40	50	60
	70	80	90	100	110	120
g519-1.pep	KEIPLDVPSQVCITRDNTQLTVDGIIYFQVTD PKLASYGSSNYIMAITQLAQTTLRSVIG					
m519-1	KEIPLDVPSQVCITRDNTQLTVDGIIYFQVTD PKLASYGSSNYIMAITQLAQTTLRSVIG					
	70	80	90	100	110	120
	130	140	150	160	170	180
g519-1.pep	RMELDKTFEERDEINSTVVSALDEAAGAWGVKVLRYEIKDLVPPQEILRAMQAQITAERE					
	:					
m519-1	RMELDKTFEERDEINSTVVAALDEAAGAWGVKVLRYEIKDLVPPQEILRSMQAQITAERE					
	130	140	150	160	170	180
	190	200	210	220	230	240
g519-1.pep	KRARIAESEGRKIEQINLASGQREAEIQQSEGEAQAAVNASNAEKIARINRAKGEAESLR					
m519-1	KRARIAESEGRKIEQINLASGQREAEIQQSEGEAQAAVNASNAEKIARINRAKGEAESLR					
	190	200	210	220	230	240
	250	260	270	280	290	300
g519-1.pep	LVAEANAIAIRQIAAALQTQGGADAVNLKIAEQYVAAFNNLAKESNTLIMPANVADIGSL					
m519-1	LVAEANAIAIRQIAAALQTQGGADAVNLKIAEQYVAAFNNLAKESNTLIMPANVADIGSL					
	250	260	270	280	290	300
	310					
g519-1.pep	ISAGMKIIDSSKTAKX					
m519-1	ISAGMKIIDSSKTAKX					
	310					

The following DNA sequence was identified in *N. meningitidis* <SEQ ID 18>:

a519-1.seq

1	ATGGAATTTT	TCATTATCTT	GCTGGCAGCC	GTCGTTGTTT	TCGGCTTCAA
51	ATCCTTTGTT	GTCATCCCAC	AGCAGGAAGT	CCACGTTGTC	GAAAGGCTCG
101	GGCGTTTCCA	TCGCGCCCTG	ACGGCCGTTT	TGAATATTTT	GATTCCCTTT
151	ATCGACCGCG	TCGCCTACCG	CCATTTCGCTG	AAAGAAATCC	CTTTAGACGT
201	ACCCAGCCAG	GTCTGCATCA	CGCGCGACAA	TACGCAGCTG	ACTGTTGACG
251	GTATCATCTA	TTTCCAAGTA	ACCGACCCCA	AACTCGCCTC	ATACGGTTCG
301	AGCAACTACA	TTATGGCGAT	TACCCAGCTT	GCCCAAACGA	CGCTGCGTTC
351	CGTTATCGGG	CGTATGGAAT	TGGACAAAAC	GTTTGAAGAA	CGCGACGAAA
401	TCAACAGCAC	CGTCGTCTCC	GCCCTCGATG	AAGCCGCCGG	AGCTTGGGGT
451	GTGAAGGTTT	TGCGTTATGA	GATTAAAGAC	TTGGTTCCGC	CGCAAGAAAT
501	CGTTCGCTCA	ATGCAGGCGC	AAATTACTGC	TGAACGCGAA	AAACGCGCCC
551	GTATCGCCGA	ATCCGAAGGT	CGTAAAATCG	AACAAATCAA	CCTTGCCAGT
601	GGTCAGCGCG	AAGCCGAAAT	CCAACAATCC	GAAGGCGAGG	CTCAGGCTGC
651	GGTCAATGCG	TCAAATGCCG	AGAAAATCGC	CCGCATCAAC	CGCGCCAAAG
701	GTGAAGCGGA	ATCCTTGCGC	CTTGTTGCCG	AAGCCAATGC	CGAAGCCATC
751	CGTCAAAATG	CCGCCGCCCT	TCAAACCCAA	GGCGGTGCGG	ATGCGGTCAA
801	TCTGAAGATT	GCGGAACAAT	ACGTCGCCGC	GTTCAACAAT	CTTGCCAAAG
851	AAAGCAATAC	GCTGATTATG	CCCGCCAATG	TTGCCGACAT	CGGCAGCCTG
901	ATTTCTGCCG	GTATGAAAAT	TATCGACAGC	AGCAAAACCG	CCAAATAA

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This corresponds to the amino acid sequence <SEQ ID 19; ORF 519-1.a>:

a519-1.pep.

```

1  MEFFFIILLAA VVVFQKSFV VIPQQEVHV ERLGRFHRAL TAGLNILIPF
51  IDRVAYRHSL KEIPLDVPSQ VCITRDNTQL TVDGIIFYQV TDPKLASYGS
101 SNYIMAITQL AQTTLRSVIG RMELDKTFEE RDEINSTVVS ALDEAAGAWG
151 VKVLRYEIKD LVPPQEILRS MQAQITAERE KRARIAESEG RKIEQINLAS
201 GQREAEIQQS EGEAQAAVNA SNAEKIARIN RAKGEAESLR LVAEANAEAI
251 RQIAAALQTQ GGADAVNLKI AEQYVAAFNN LAKESNTLIM PANVADIGSL
301 ISAGMKIIDS SKTAK*
```

m519-1/a519-1 ORFs 519-1 and 519-1.a showed a 99.0% identity in 315 aa overlap

a519-1.pep	10	20	30	40	50	60
	MEFFFIILLAAVVVFQKSFVVIPQQEVHVVERLGRFHRALTAGLNILIPFIDRVAYRHSL					
m519-1	MEFFFIILLVAVAVVFQKSFVVIPQQEVHVVERLGRFHRALTAGLNILIPFIDRVAYRHSL					
	10	20	30	40	50	60
a519-1.pep	70	80	90	100	110	120
	KEIPLDVPSQVCITRDNTQLTVDGIIFYQVTDPKLASYGSSNYIMAITQLAQTTLRSVIG					
m519-1	KEIPLDVPSQVCITRDNTQLTVDGIIFYQVTDPKLASYGSSNYIMAITQLAQTTLRSVIG					
	70	80	90	100	110	120
a519-1.pep	130	140	150	160	170	180
	RMELDKTFEERDEINSTVVSALDEAAGAWGVKVLRYEIKDLVPPQEILRSMQQAITAERE					
m519-1	RMELDKTFEERDEINSTVVAALDEAAGAWGVKVLRYEIKDLVPPQEILRSMQQAITAERE					
	130	140	150	160	170	180
a519-1.pep	190	200	210	220	230	240
	KRARIAESEGRKIEQINLASGQREAEIQQSEGEAQAAVNASNAEKIARINRAKGEAESLR					
m519-1	KRARIAESEGRKIEQINLASGQREAEIQQSEGEAQAAVNASNAEKIARINRAKGEAESLR					
	190	200	210	220	230	240
a519-1.pep	250	260	270	280	290	300
	LVAEANAEAIRQIAAALQTQGGADAVNLKIAEQYVAAFNNLAKESNTLIMPANVADIGSL					
m519-1	LVAEANAEAIRQIAAALQTQGGADAVNLKIAEQYVAAFNNLAKESNTLIMPANVADIGSL					
	250	260	270	280	290	300
a519-1.pep	310					
	ISAGMKIIDSSKTAKX					
m519-1	ISAGMKIIDSSKTAKX					
	310					

576 and 576-1 gnm22.seq

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 20>:

m576.seq.. (partial)

```

1  ..ATGCAGCAGG CAAGCTATGC GATGGGCGTG GACATCGGAC GCTCCCTGAA
51  GCAAATGAAG GAACAGGGCG CGGAAATCGA TTTGAAAGTC TTTACCGAAG
101 CCATGCAGGC AGTGTATGAC GGCAAAGAAA TCAAATGAC CGAAGAGCAG
151 GCTCAGGAAG TCATGATGAA ATTCCTTCAG GAACAACAGG CTAAAGCCGT
201 AGAAAAACAC AAGGCGGACG CGAAGGCCAA TAAAGAAAAA GGCGAAGCCT
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251	TTCTGAAAGA	AAATGCCGCC	AAAGACGGCG	TGAAGACCAC	TGCTTCCGGC
301	CTGCAATACA	AAATCACCAA	ACAGGGCGAA	GGCAAACAGC	CGACAAAGA
351	CGACATCGTT	ACCGTGGAAT	ACGAAGGCCG	CCTGATTGAC	GTTACCGTAT
401	TCGACAGCAG	CAAAGCCAA	GCGCGCCCGG	TACACTTCCC	TTTGAGCCAA
451	GTGATTCCGG	GTTGGACCGA	AGCGGTACAG	CTTCTGAAAG	AAGGCGGCGA
501	AGCCACGTTT	TACATCCCGT	CCAACCTTGC	CTACCGCGAA	CAGGGTGCGG
551	GCGCAACAAAT	CGGTCCGAAC	GCCACTTTGG	TATTTGATGT	GAAACTGGTC
601	AAAATCGGCG	CACCCGAAAA	GCGCCCCGCC	AAGCAGCCGG	CTCAAGTCGA
651	CATCAAAAAA	GTAATTAA			

This corresponds to the amino acid sequence <SEQ ID 21; ORF 576>:

m576.pep.. (partial)

1	..MQQASYAMGV	DIGRSLKQMK	EQGAEIDLKV	FTEAMQAVYD	GKEIKMTEEQ
51	AQEVMMKFQ	EQQAQAVEKH	KADAKANKEK	GEAFLKENAA	KDGVKTTASG
101	LQYKITKQGV	GKQPTKKDDIV	TVEYEGRLID	GTVFDSSKAN	GGPVTFFPLSQ
151	VIPGWTEQGV	LLKEGGEATF	YIPSNLAYRE	QGAGDKIGPN	ATLVFVDVKLV
201	KIGAPENAPA	KQPAQVDIKK	VN*		

The following partial DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 22>:

g576.seq.. (partial)

1	..atgggcgtgg	acatcggacg	ctccctgaaa	caaatgaagg	aacagggcgc
51	ggaaatcgat	ttgaaagtct	ttacgcgatg	catgcaggca	gtgtatgacg
101	gcaaaagaaat	caaaaatgac	gaagagcagg	cccaggaagt	gatgatgaaa
151	ttcctgcagg	agcagcaggc	taaagccgta	gaaaaacaca	aggcggatgc
201	gaaggccaac	aaagaaaaag	gcgaagcctt	cctgaaggaa	aatgccgcgc
251	aagacggcgt	gaagaccact	gcttcgggtc	tgcagtacaa	aatcaccaaa
301	cagggtgaag	gcaaacagcc	gacaaaagac	gacatcgtaa	ccgtggaata
351	cgaaggccgc	ctgattgacg	gtaccgtatt	cgacagcagc	aaagccaacg
401	gcggcccgcc	caccttccct	ttgagccaag	tgattccggg	ttggaccgaa
451	ggcgtacggc	ttctgaaaga	aggcggcgaa	gccacgttct	acatcccgtc
501	caacctttgcc	taccgcgaag	agggtgcggg	cgaaaaaatc	ggtcgaagac
551	ccacttttgt	atttgacgtg	aaacttgtca	aaatcggcgc	acccgaaaaac
601	gcgcccgcga	agcagccgga	tcaagtcgac	atcaaaaaag	taaattaa

This corresponds to the amino acid sequence <SEQ ID 23; ORF 576.ng>:

g576.pep..(partial)

1	MGVDIGRSLK	QMKEQGAIED	LKVFTDAMQA	VYDGKEIKMT	EEQAQEVMMK
51	FLQEQQAKAV	EKKHADAKAN	KEKGAEFLKE	NAAEDGVKTT	ASGLQYKITK
101	QGEGKQPTKD	DIVTVEYEGR	LIDGTVFDSS	KANGGPATFP	LSQVIPGWTE
151	GVRLLEKGE	ATFYIPSNLA	YREQGAGEKI	GPNATLVFDV	KLKVIKAPEN
201	APAKQPDQVD	IKKVN*			

Computer analysis of this amino acid sequence gave the following results:

Homology with a predicted ORF from *N. gonorrhoeae*

m576/g576 97.2% identity in 215 aa overlap

	10	20	30	40	50	60
m576.pep	MQQASYAMGVDIGRSLKQMKEQGAIEDLKVFTEAMQAVYDGKEIKMTEEQAQEVMMKFLQ					
g576	:					
	10	20	30	40	50	
	MGVDIGRSLKQMKEQGAIEDLKVFTEAMQAVYDGKEIKMTEEQAQEVMMKFLQ					
	70	80	90	100	110	120
m576.pep	EQQAKAVEKHKADAKANKEKGEAFLKENAAKDGVKTTASGLQYKITKQEGEKQPTKDDIV					
g576	:					
	60	70	80	90	100	110
	EQQAKAVEKHKADAKANKEKGEAFLKENAAEDGVKTTASGLQYKITKQEGEKQPTKDDIV					

	130	140	150	160	170	180
m576.pep	TVEYEGRLIDGTVFDS	SKANGGPVTFPLSQV	IPGWTEGVQLLKEG	GEATFYIPSNLAYRE		
g576	TVEYEGRLIDGTVFDS	SKANGGPATFPLSQV	IPGWTEGVRLLEKG	GEATFYIPSNLAYRE		
	120	130	140	150	160	170
	190	200	210	220		
m576.pep	QGAGDKIGPNATLVF	DVKLVKIGAPENAPAK	QPAQVDIIKKVN	X		
g576	QGAGEKIGPNATLVF	DVKLVKIGAPENAPAK	QPDQVDIIKKVN	X		
	180	190	200	210		

```
a576.seq
1  ATGAACACCA TTTTCAA AAT CAGCGCA CTG ACCCTTT CCG CCGCTTT GGC
51  ACTTTCCGCC TGCCTCA AAAA AAGAAGCC GC CCCCGCAT CT GCATCCG AAC
101 CTGCCGCCGC TTGTTCC GCG CAGGGCG ACA CCTTTCG AT CGGCAGC AGC
151 ATGCAGCAGG CAAGCTAT GC GATGGGCG TG GACATCGG AC GCTCCCTG AA
201 GCAAATGAAG GAACAGG GCG CGGAAAT CGA TTTGAAAG TC TTTACCGA AG
251 CCATGCAGGC AGTGTATG AC GGCAAAGAAA TCAAAATG AC CGAAGAGC AG
301 GCTCAGGAAG TCATGTAG AA ATTCCTT CAG GAACAACAG G CTAAGGCC GT
351 AGAAAAACAC AAGGCGG ACG CGAAGGCC AA TAAAGAAAAA GCGGAAGC CT
401 TTCTGAAAGA AAATGCC GCC AAAGACGG CG TGAAGACC AC TGCTTCCG GC
451 CTGCAATACA AAATACCCA ACACAGGG CGAA GGCAAAAC AG CGACCAAA GA
501 CGACATCGTT ACCGTGGA AT ACGAAGGCC G CCTGATTG AC GGTACGGT AT
551 TCGACAGCAG CAAGAAGC AAC GGCGCCCG CG TCACTTCCC C TTTGAGCC AA
601 GTGATTCTGG GTTGGAACC GA AGGCGTAC AG CTTCTGAA AG AAGGCGGC GA
651 AGCCAGGTTT CACATCCC GT CCAACCTT GC CTACCGCG AA CAGGGTGC GG
701 GCGACAAAAA CGGCCGGA AC GCCACTTT GG TATTTGAT GT GAAACTGG TC
751 AAAATCGGCG CACCCGAAA ACGCGCCCG CC AAGCAGCC GG CTC AAGTCGA
801 CATCAAAAAA GTAAATTA A
```

a576.pep

1	MNTIFKISAL	<u>TL</u> SAALALSA	CGKKEAAPAS	ASEPAAASSA	QGDTSIGST
51	MQQASYAMGV	DIGRSLKQMK	EQGAEIDLKV	FTEAMQAVYD	GKEIKMTEEQ
101	AQEVMMKFLQ	EQQAKAVEKH	KADAKANKEK	GEAFLKENAA	KDGVKTTASG
151	LQYKITKQGE	GKQPTKDDIV	TVEYEGRLID	GTVFDSSKAN	GGPVTFFPLSQ
201	VILGWTEGVQ	LLKEQGEATF	YIPSNLAYRE	QGAGDKIGPN	ATLVFDFVKLV
251	KIGAPENAPA	KQPAQVDIIK	VN*		

[illegible]

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	160	170	180	190	200	210
m576.pep	VIPGWTEGVQLLKEGGEATFYIPSNLAYREQGAGDKIGPNATLVFDVKLVKIGAPENAPA					
a576	VILGWTEGVQLLKEGGEATFYIPSNLAYREQGAGDKIGPNATLVFDVKLVKIGAPENAPA					
	210	220	230	240	250	260
		220				
m576.pep	KQPAQVDIKKVN					
a576	KQPAQVDIKKVN					
		270				

Further work revealed the following DNA sequence identified in *N. meningitidis* <SEQ ID 26>:

```

m576-1.seq
1  ATGAACACCA TTTTCAAAT CAGCGCACTG ACCCTTTCCG CCGCTTTGGC
51  ACTTTCCGCC TGC GGCAAAA AAGAAGCCGC CCCCGCATCT GCATCCGAAC
101 CTGCCGCCGC TTCTCCGCG CAGGGCGACA CCTCTCGAT CGGCAGCAGC
151 ATGCAGCAGG CAAGCTATGC GATGGGCGTG GACATCGGAC GCTCCCTGAA
201 GCAAATGAAG GAACAGGGCG CGGAAATCGA TTTGAAAGTC TTTACCGAAG
251 CCATGCAGGC AGTGTATGAC GGCAAAGAAA TCAAATGAC CGAAGAGCAG
301 GCTCAGGAAG TCATGATGAA ATTCTTCAG GAACAACAGG CTAAAGCCGT
351 AGAAAAACAC AAGGCGGACG CGAAGGCCAA TAAAGAAAAA GGCGAAGCCT
401 TTCTGAAAGA AAATGCCGCC AAAGACGGCG TGAAGACCAC TGCTTCCGGC
451 CTGCAATACA AAATCACCAC ACAGGGCGAA GGCAAACAGC CGACCAAAGA
501 CGACATCGTT ACCGTGGAAT ACGAAGGCCG CCTGATTGAC GGTACGGTAT
551 TCGACAGCAG CAAAGCCAAC GCGGCGCCCG TCACCTTCCC TTTGAGCCAA
601 GTGATTCCGG GTTGGACCGA AGGCGTACAG CTTCTGAAAG AAGGCGGCGA
651 AGCCACGTTT TACATCCCGT CCAACCTTGC CTACCGCGAA CAGGGTGCGG
701 GCGACAAAAT CGGTCCGAAC GCCACTTTGG TATTTGATGT GAAACTGGTC
751 AAAATCGGCG CACCCGAAAA CGCGCCCGCC AAGCAGCCGG CTCAAGTCGA
801 CATCAAAAAA GTAAATTAA

```

This corresponds to the amino acid sequence <SEQ ID 27; ORF 576-1>:

```

m576-1.pep
1  MNTIFKISAL TLSAALALSA CGKKEAAPAS ASEPAASSA QGDTSSIGST
51  MQQASYAMGV DIGRSLKQMK EQGAEIDLKV FTEAMQAVYD GKEIKMTEEQ
101 AQEVMKFLQ EQQAKAVEKH KADAKANKEK GEAFLENAA KDGVKTTASG
151 LQYKITKQGE GKQPTKDDIV TVEYEGRLID GTVFDSSKAN GGPVTFPLSQ
201 VIPGWTEGVQ LLKEGGEATF YIPSNLAYRE QGAGDKIGPN ATLVFDVKLV
251 KIGAPENAPA KQPAQVDIKK VN*

```

The following DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 28>:

```

g576-1.seq
1  ATGAACACCA TTTTCAAAT CAGCGCACTG ACCCTTTCCG CCGCTTTGGC
51  ACTTTCCGCC TGC GGCAAAA AAGAAGCCGC CCCCGCATCT GCATCCGAAC
101 CTGCCGCCGC TTCTGCCGCG CAGGGCGACA CCTCTCAAT CGGCAGCAGC
151 ATGCAGCAGG CAAGCTATGC AATGGGCGTG GACATCGGAC GCTCCCTGAA
201 ACAAATGAAG GAACAGGGCG CGGAAATCGA TTTGAAAGTC TTTACCGATG
251 CCATGCAGGC AGTGTATGAC GGCAAAGAAA TCAAATGAC CGAAGAGCAG
301 GCCCAGGAAG TGATGATGAA ATTCTGCGAG GAGCAGCAGG CTAAAGCCGT
351 AGAAAAACAC AAGGCGGATG CGAAGGCCAA CAAAGAAAAA GGCGAAGCCT
401 TCCTGAAGGA AAATGCCGCC AAAGACGGCG TGAAGACCAC TGCTTCCGGT
451 CTGCAGTACA AAATCACCAC ACAGGGTGAA GGCAAACAGC CGACCAAAGA
501 CGACATCGTT ACCGTGGAAT ACGAAGGCCG CCTGATTGAC GGTACGGTAT
551 TCGACAGCAG CAAAGCCAAC GCGGCGCCCG CCACCTTCCC TTTGAGCCAA
601 GTGATTCCGG GTTGGACCGA AGGCGTACGG CTTCTGAAAG AAGGCGGCGA
651 AGCCACGTTT TACATCCCGT CCAACCTTGC CTACCGCGAA CAGGGTGCGG
701 GCGAAAAAAT CGGTCCGAAC GCCACTTTGG TATTTGACGT GAAACTGGTC
751 AAAATCGGCG CACCCGAAAA CGCGCCCGCC AAGCAGCCGG ATCAAGTCGA

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801 CATCAAAAAA GTAAATTAA

This corresponds to the amino acid sequence <SEQ ID 29; ORF 576-1.ng>:

g576-1.pep

```

1  MNTIFKISAL TLSAALALSA CGKKEAAPAS ASEPAASAA QGDTSSIGST
51  MQQASYAMGV DIGRSLKQMK EQGAEIDLKV FTDAMQAVYD GKEIKMTEEQ
101 AQEVMKFLQ EQQAKAVEKH KADAKANKEK GEAFLENAA KDGVKTTASG
151 LQYKITKQGE GKQPTKDDIV TVEYEGRLID GTVFDSSKAN GGPATFPLSQ
201 VIPGWTEGVR LLKEGGEATF YIPSNLAYRE QGAGEKIGPN ATLVFDVKLV
251 KIGAPENAPA KQPDQVDIKK VN*

```

g576-1/m576-1 ORFs 576-1 and 576-1.ng showed a 97.8% identity in 272 aa overlap

	10	20	30	40	50	60
g576-1.pep	MNTIFKISAL	TLSAALALSA	CGKKEAAPAS	ASEPAASAA	QGDTSSIGST	MQQASYAMGV
m576-1	MNTIFKISAL	TLSAALALSA	CGKKEAAPAS	ASEPAASSA	QGDTSSIGST	MQQASYAMGV
	10	20	30	40	50	60
g576-1.pep	70	80	90	100	110	120
	DIGRSLKQMK	EQGAEIDLKV	FTDAMQAVYD	GKEIKMTEEQ	AQEVMKFLQ	EQQAKAVEKH
m576-1	DIGRSLKQMK	EQGAEIDLKV	FTDAMQAVYD	GKEIKMTEEQ	AQEVMKFLQ	EQQAKAVEKH
	70	80	90	100	110	120
g576-1.pep	130	140	150	160	170	180
	KADAKANKEK	GEAFLENAA	KDGVKTTASG	LQYKITKQGE	GKQPTKDDIV	TVEYEGRLID
m576-1	KADAKANKEK	GEAFLENAA	KDGVKTTASG	LQYKITKQGE	GKQPTKDDIV	TVEYEGRLID
	130	140	150	160	170	180
g576-1.pep	190	200	210	220	230	240
	GTVDSSKAN	GGPATFPLSQ	VIPGWTEGVR	LLKEGGEATF	YIPSNLAYRE	QGAGEKIGPN
m576-1	GTVDSSKAN	GGPATFPLSQ	VIPGWTEGVR	LLKEGGEATF	YIPSNLAYRE	QGAGEKIGPN
	190	200	210	220	230	240
g576-1.pep	250	260	270			
	ATLVFDVKLV	KIGAPENAPA	KQPDQVDIKK	VN		
m576-1	ATLVFDVKLV	KIGAPENAPA	KQPDQVDIKK	VN		
	250	260	270			

The following DNA sequence was identified in *N. meningitidis* <SEQ ID 30>:

a576-1.seq

```

1  ATGAACACCA TTTTCAAAAT CAGCGCACTG ACCCTTTCCG CCGCTTTGGC
51  ACTTTCCGCC TCGGCAAAA AAGAAGCCGC CCCCGCATCT GCATCCGAAC
101 CTGCCGCCGC TTCTTCCGCG CAGGGCGACA CCTCTTCGAT CGGCAGCAGC
151 ATGCAGCAGG CAAGCTATGC GATGGGCGTG GACATCGGAC GCTCCCTGAA
201 GCAAATGAAG GAACAGGGCG CGGAAATCGA TTTGAAAGTC TTTACCGAAG
251 CCTATGCAGG AGTGTATGAC GGCAAAGAAA TCAAATGAC CGAAGAGCAG
301 GCTCAGGAAG TCATGATGAA ATTCCTTCAG GAACAACAGG CTAAGCCGT
351 AGAAAAACAC AAGGCGGACG CGAAGGCCAA TAAAGAAAAA GGCGAAGCCT
401 TTCTGAAAGA AAATGCCGCC AAAGACGGCG TGAAGACCAC TGCTTCCGGC
451 CTGCAATACA AAATCACCAA ACAGGGCGAA GGCAAACAGC CGACCAAAGA
501 CGACATCGTT ACCGTGGAAT ACGAAGGCCG CCTGATTGAC GGTACGGTAT
551 TCGACAGCAG CAAAGCCAAC GGCGGCCCGG TCACCTTCCC TTTGAGCCAA
601 GTGATTCTGG GTTGGACCGA AGGCGTACAG CTTCTGAAAG AAGGCGGCGA
651 AGCCACGTTT TACATCCCGT CCAACCTTGC CTACCGCGAA CAGGGTGCGG
701 GCGACAAAAT CGGCCCAAC GCCACTTTGG TATTTGATGT GAACTGCTC

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751 AAAATCGGCG CACCCGAAAA CGCGCCCGCC AAGCAGCCGG CTCAAGTCGA
 801 CATCAAAAAA GTAAATTAA

This corresponds to the amino acid sequence <SEQ ID 31; ORF 576-1.a>:

a576-1.pep
 1 MNTIFKISAL TLSAALALSA CGKKEAAPAS ASEPAASSA QGDTSSIGST
 51 MQQASYAMGV DIGRSLKQMK EQGAEIDLKV FTEAMQAVYD GKEIKMTEEQ
 101 AQEVMMKFLQ EQQAKAVEKH KADAKANKEK GEAFLENAA KDGVKTTASG
 151 LQYKITKQGE GKQPTKDDIV TVEYEGRLID GTVFDSSKAN GGPVTFPLSQ
 201 VILGWTEGVQ LLKEGGEATF YIPSNLAYRE QGAGDKIGPN ATLVFDVKLV
 251 KIGAPENAPA KQPAQVDIKK VN*

a576-1/m576-1 ORFs 576-1 and 576-1.a 99.6% identity in 272 aa overlap

	10	20	30	40	50	60
a576-1.pep	MNTIFKISAL	TLSAALALS	SACGKKEA	PASASEPA	ASSAQGDT	SSIGSTMQ
m576-1	MNTIFKISAL	TLSAALALS	SACGKKEA	PASASEPA	ASSAQGDT	SSIGSTMQ
	10	20	30	40	50	60
	70	80	90	100	110	120
a576-1.pep	DIGRSLKQ	MKEQGA	EIDLKVF	TEAMQAV	YDGKEIK	MTEEQAE
m576-1	DIGRSLKQ	MKEQGA	EIDLKVF	TEAMQAV	YDGKEIK	MTEEQAE
	70	80	90	100	110	120
	130	140	150	160	170	180
a576-1.pep	KADAKANKE	KGEAFLK	ENAAKDG	VKT TAS	GLQYKIT	KQGEKG
m576-1	KADAKANKE	KGEAFLK	ENAAKDG	VKT TAS	GLQYKIT	KQGEKG
	130	140	150	160	170	180
	190	200	210	220	230	240
a576-1.pep	GT VFDSS	KANGGP	VT FPLS	QVILGW	TEGVQL	LKEGGE
m576-1	GT VFDSS	KANGGP	VT FPLS	QVILGW	TEGVQL	LKEGGE
	190	200	210	220	230	240
	250	260	270			
a576-1.pep	ATLVFDV	KL VKI	GAPENAP	AKQPAQ	VDIKK	VNX
m576-1	ATLVFDV	KL VKI	GAPENAP	AKQPAQ	VDIKK	VNX
	250	260	270			

919 and 919-2 gnm43.seq

The following partial DNA sequence was identified in *N.meningitidis* <SEQ ID 32>:

m919.seq
 1 ATGAAAAAAT ACCTATTCCG CGCCGCCCTG TACGGCATCG CCGCCGCCAT
 51 CCTCGCCGCC TGCCAAAGCA AGAGCATCCA AACCTTTCCG CAACCCGACA
 101 CATCCGTCAT CAACGGCCCG GACCGGCCGG TCGGCATCCC CGACCCCGCC
 151 GGAACGACGG TCGGCGGCGG CGGGGCCGTC TATACCGTTG TACCGCACCT
 201 GTCCCTGCCC CACTGGGCGG CGCAGGATTT CGCCAAAAGC CTGCAATCCT
 251 TCCGCCTCGG CTGCGCCAAT TTGAAAAACC GCCAAGGCTG GCAGGATGTG
 301 TGCGCCCAAG CCTTTCAAAC CCCCGTCCAT TCCTTTCAGG CAAAACAGTT
 351 TTTTGAACGC TATTTACAGC CGTGGCAGGT TGCAGGCAAC GGAAGCCTTG

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401 CCGGTACGGT TACCGGCTAT TACGAACCGG TGCTGAAGGG CGACGACAGG
451 CGGACGGCAC AAGCCCGCTT CCCGATTTCAC GGTATTCCTG ACGATTTTAT
501 CTCCGTCCCC CTGCCTGCCG GTTTGCGGAG CGGAAAAGCC CTTGTCCGCA
551 TCAGGCAGAC GGGAAAAAAC AGCGGCACAA TCGACAATAC CGGCGGCACA
601 CATACCGCCG ACCTCTCCCG ATTCCCACAT ACCGCGCGCA CAACAGCAAT
651 CAAAGGCAGG TTTGAAGGAA GCCGCTTCCT CCCCTACCAC ACGCGCAACC
701 AAATCAACGG CGGCGCGCTT GACGGCAAAG CCCCATACT CCGTTACGCC
751 GAAGACCCCTG TCGAACTTTT TTTTATGCAC ATCCAAGGCT CGGGCCGTCT
801 GAAAACCCCG TCCGGCAAAT ACATCCGCAT CGGCTATGCC GACAAAAACG
851 AACATCCyTA CGTTTCCATC GGACGCTATA TGGCGGATAA GGGCTACCTC
901 AAACCTCGAC AAACCTCCAT GCAGGGCATT AAGTCTTATA TCGCGCAAAA
951 TCCGCAACGC CTCGCCGAAG TTTTGGGTCA AAACCCAGC TATATCTTTT
1001 TCCGCGAGCT TGCCGGAAGC AGCAATGACG GCCCTGTCCG CGCACTGGGC
1051 ACGCCGCTGA TGGGGGAATA TGCCGCGCGCA GTCGACCGGC ACTACATTAC
1101 CTTGGGTGCG CCCTTATTTG TCGCCACCGC CCATCCGGTT ACCCGCAAAG
1151 CCCTCAACCG CCTGATTATG GCGCAGGATA CCGGCAGCGC GATTAAAGGC
1201 GCGGTGCGCG TGGATTATTT TTGGGGATAC GCGCAGCAAG CCGGCGAACT
1251 TGCCGGCAAA CAGAAAACCA CGGGATATGT CTGGCAGCTC CTACCCAACG
1301 GTATGAAGCC CGAATACCGC CCGTAA

```

This corresponds to the amino acid sequence <SEQ ID 33; ORF 919>:

m919.pep

```

1 MKKYLFRAL YGIAAAILAA CQSKSIQTFP QPDTSVINGP DRPVGIPDPA
51 GTTVGGGGAV YTVVPHLSLP HWAAQDFAKS LQSFRLGCAN LKNRQGWQDV
101 CAQAFQTPVH SFQAKQFFER YFTPWQVAGN GSLAGTVTGY YEPVLKGDDR
151 RTAQARFPIY GIPDDFISVP LPAGLRSGKA LVRI RQTGKN SGTIDNTGGT
201 HTADLSRFPI TARTTAIKGR FEGRSFLPYH TRNQINGGAL DGKAPILGYA
251 EDPVELFFMH IQSGRLKTP SGKYIRIGYA DKNEHPYVSI GRYMADKGYL
301 KLGQTSMQGI KSYMQRNPQR LAEVLGQNPS YIFFRELAYS SNGDPVAGLG
351 TPLMGEYAGA VDRHYITLGA PLFVATAHPV TRKALNRLIM AQDTGSAIKG
401 AVRVDYFWGY GDEAGELAGK QKTTGYVWQL LPNGMKPEYR P*

```

The following partial DNA sequence was identified in *N.meningitidis* <SEQ ID 34>:

m919-2.seq

```

1 ATGAAAAAAT ACCTATTCCG CGCGGCCCTG TACGGCATCG CCGCCGCCAT
51 CCTCGCCGCC TGCCAAAGCA AGAGCATCCA AACCTTTCCG CAACCCGACA
101 CATCCGTCAT CAACGGCCCC GACCGGCCGG TCGGCATCCC CGACCCCGCC
151 GGAACGACGG TCGGCGGCGG CGGGGCCGTC TATACCGTTG TACCGCACTT
201 GTCCCTGCCC CACTGGGCGG CGCAGGATTT CGCCAAAAGC CTGCAATCCT
251 TCCGCCTCGG CTGCGCCAAT TTGAAAACC GCCAAGGCTG GCAGGATGTG
301 TGCGCCCAAG CCTTTCAAAC CCCCGTCCAT TCCTTTCAGG CAAAACAGTT
351 TTTTGAACGC TATTTACGC CGTGGCAGGT TGCAGGCAAC GGAAGCCTTG
401 CCGGTACGGT TACCGGCTAT TACGAACCGG TGCTGAAGGG CGACGACAGG
451 CGGACGGCAC AAGCCCGCTT CCCGATTTCAC GGTATTCCTG ACGATTTTAT
501 CTCCGTCCCC CTGCCTGCCG GTTTGCGGAG CGGAAAAGCC CTTGTCCGCA
551 TCAGGCAGAC GGGAAAAAAC AGCGGCACAA TCGACAATAC CGGCGGCACA
601 CATACCGCCG ACCTCTCCCG ATTCCCACAT ACCGCGCGCA CAACAGCAAT
651 CAAAGGCAGG TTTGAAGGAA GCCGCTTCCT CCCCTACCAC ACGCGCAACC
701 AAATCAACGG CGGCGCGCTT GACGGCAAAG CCCCATACT CCGTTACGCC
751 GAAGACCCCTG TCGAACTTTT TTTTATGCAC ATCCAAGGCT CGGGCCGTCT
801 GAAAACCCCG TCCGGCAAAT ACATCCGCAT CGGCTATGCC GACAAAAACG
851 AACATCCCTA CGTTTCCATC GGACGCTATA TGGCGGATAA GGGCTACCTC
901 AAACCTCGAC AAACCTCCAT GCAGGGCATT AAGTCTTATA TCGCGCAAAA
951 TCCGCAACGC CTCGCCGAAG TTTTGGGTCA AAACCCAGC TATATCTTTT
1001 TCCGCGAGCT TGCCGGAAGC AGCAATGACG GCCCTGTCCG CGCACTGGGC
1051 ACGCCGCTGA TGGGGGAATA TGCCGCGCGCA GTCGACCGGC ACTACATTAC
1101 CTTGGGTGCG CCCTTATTTG TCGCCACCGC CCATCCGGTT ACCCGCAAAG
1151 CCCTCAACCG CCTGATTATG GCGCAGGATA CCGGCAGCGC GATTAAAGGC

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1201 GCGGTGCGCG TGGATTATTT TTGGGGATAC GGCGACGAAG CCGGCGAACT
1251 TGCCGGCAAA CAGAAAACCA CGGGATATGT CTGGCAGCTC CTACCCAACG
1301 GTATGAAGCC CGAATACCGC CCGTAA

```

This corresponds to the amino acid sequence <SEQ ID 35; ORF 919-2>:

m919-2.pep

```

1 MKKYLFRAL YGIAAAILAA CQSKSIQTFP QPDTSVINGP DRPVGIPDPA
51 GTTVGGGGAV YTVVPHLSLP HWAAQDFAKS LQSFRLGCAN LKNRQGWQDV
101 CAQAFQTPVH SFQAKQFFER YFTPWQVAGN GSLAGTVTGY YEPVLKGDGR
151 RTAQARFPIY GIPDDFISVP LPAGLRSGKA LVRIQRTGKN SGTIDNTGGT
201 HTADLSRFPI TARTTAIKGR FEGRSFLPYH TRNQINGGAL DGKAPILGYA
251 EDPVELFFMH IQGSGRLKTP SGKYIRIGYA DKNEHPYVSI GRYMADKGYL
301 KLGQTSMQGI KSYMQRNPQR LAEVLGQNPS YIFFRELAYS SNDGPVAGLG
351 TPLMGEYAGA VDRHYITLGA PLFVATAHPV TRKALNRLIM AQDTGSAIKG
401 AVRVDYFWGY GDEAGELAGK QKTTGYVWQL LPNGMKPEYR P*

```

The following partial DNA sequence was identified in *N.gonorrhoeae* <SEQ ID 36>:

g919.seq

```

1 ATGAAAAAAC ACCTGCTCCG CTCCGCCCTG TACGGcatCG CCGCCgccAT
51 CctcgCCGCC TGCCAAAgca gGAGCATCCA AACCTTTCCG CAACCCGACA
101 CATCCGTCAT CAACGGCCCG GACCGGCCGG CCGGCATCCC CGACCCCGCC
151 GGAACGACGG TTGCCGGCGG CGGGGCCGTC TATACCGTTG TGCCGCACCT
201 GTCCATGCCC CACTGGGCGG CGCaggATTT TGCCAAAAGC CTGCAATCCT
251 TCCGCCTCGG CTGCGCCAAT TTGAAAAACC GCCAAGGCTG GCAGGATGTG
301 TGCGCCCAAG CCTTTCAAAC CCCCCTGCAT TCCTTTCAGG CAAAGcGgTT
351 TTTTGAACGC TATTTACAGC cgtGGCaggT tgcaggcaAC GGAAGcCTTG
401 Caggtacggt TACCGGCTAT TACGAACCGG TGCTGAAGGG CGACGGCAGG
451 CGGACGGAAC GGGCCCGCTT CCCGATTTAC GGTATTCCCG ACGATTTTAT
501 CTCCGTCCCG CTGCCTGCCG GTTTGCGGGG CGGAAAAAAC CTTGTCCGCA
551 TCAGGCAGAc ggGAAAAAAC AGCGGCACGA TCGACAATGC CGGCGGCACG
601 CATAACGCCC ACCTCTCCCG ATTCCCCATC ACCGCGCGCA CAACGGcaat
651 caaaGGCAGG TTTGAaggAA GCCGCTTCCT CCCTTACCAC ACGCGCAACC
701 AAAtcaacGG CGGCgcgcTT GACGGCAAag cccCCATCCT CggttacgcC
751 GAagaccCcG tccggcaaat TTTCATGCAC AtccaaggCT CGGGCCGCTT
801 GAAAACCCcg tccggcaaat acatCCGCAt cggATacgcc gacAAAAACG
851 AACAtccgTa tgtttccatc ggACGctaTA TGGCGGACAA AGGCTACCTC
901 AAGctcgggc agACCTCGAT GCAGGgcatc aaagcCTATA TGCGGCAAAA
951 TCCGCAACGC CTCGCCGAAG TTTTGGGTCA AAACCCAGC TATATCTTTT
1001 TCCGCGAGCT TGCCGGAAGC GGCAATGAGG GCCCGCTCGG CGCACTGGGC
1051 ACGCCACTGA TGGGGGAATA CGCCGGCGCA ATCGACCGGC ACTACATTAC
1101 CTTGGGCGCG CCCTTATTTG TCGCCACCGC CCATCCGTTT ACCGCAAGG
1151 CCTCAACCG CCTGATTATG GCGCAGGATA CAGGCAGCGC GATCAAAGGC
1201 GCGGTGCGCG TGGATTATTT TTGGGGTTAC GGCGACGAAG CCGGCGAACT
1251 TGCCGGCAAA CAGAAAACCA CGGGATACGT CTGGCAGCTC CTGCCCAACG
1301 GCATGAAGCC CGAATACCGC CCGTGA

```

This corresponds to the amino acid sequence <SEQ ID 37; ORF 919.ng>:

g919.pep

```

1 MKKHLLRSAL YGIAAAILAA CQSRSIQTFP QPDTSVINGP DRPAGIPDPA
51 GTTVAGGGAV YTVVPHLSMP HWAAQDFAKS LQSFRLGCAN LKNRQGWQDV
101 CAQAFQTPVH SFQAKRFFER YFTPWQVAGN GSLAGTVTGY YEPVLKGDGR
151 RTERARFPIY GIPDDFISVP LPAGLRGGKN LVRIQRTGKN SGTIDNAGGT
201 HTADLSRFPI TARTTAIKGR FEGRSFLPYH TRNQINGGAL DGKAPILGYA
251 EDPVELFFMH IQGSGRLKTP SGKYIRIGYA DKNEHPYVSI GRYMADKGYL
301 KLGQTSMQGI KAYMRNPQR LAEVLGQNPS YIFFRELAYS GNEGPVAGLG
351 TPLMGEYAGA IDRHYITLGA PLFVATAHPV TRKALNRLIM AQDTGSAIKG
401 AVRVDYFWGY GDEAGELAGK QKTTGYVWQL LPNGMKPEYR P*

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ORF 919 shows 95.9 % identity over a 441 aa overlap with a predicted ORF (ORF 919.ng) from *N. gonorrhoeae*:

m919/g919

	10	20	30	40	50	60
m919.pep	MKKYLFRAALYGIAAAAILAACQSKSIQTFFQPDTSVINGPDRPVGIPDPAGTTVGGGGAV					
	: : : : :					
g919	MKKHLLRSALYGIAAAAILAACQSRISIQTFFQPDTSVINGPDRPAGIPDPAGTTVAGGGAV					
	10	20	30	40	50	60
	70	80	90	100	110	120
m919.pep	YTVVPHLSLPHWAAQDFAKSLQSFRLGCANLKNRQGWQDVCAQAFQTPVHSFQAKQFFER					
	:					
g919	YTVVPHLSMPHWAAQDFAKSLQSFRLGCANLKNRQGWQDVCAQAFQTPVHSFQAKRFFER					
	70	80	90	100	110	120
	130	140	150	160	170	180
m919.pep	YFTPWQVAGNGSLAGTVTGYEYPVLKGDGRRTERARFPIYGI PDDFISVPLPAGLRSGKA					
	:					
g919	YFTPWQVAGNGSLAGTVTGYEYPVLKGDGRRTERARFPIYGI PDDFISVPLPAGLRGGKN					
	130	140	150	160	170	180
	190	200	210	220	230	240
m919.pep	LVRIRQTGKNSGTIDNTGGTHTADLSRFPITARTTAIKGRFEGSRFLPYHTRNQINGGAL					
	:					
g919	LVRIRQTGKNSGTIDNAGGHTHTADLSRFPITARTTAIKGRFEGSRFLPYHTRNQINGGAL					
	190	200	210	220	230	240
	250	260	270	280	290	300
m919.pep	DGKAPILGYAEDPVELFFMHIIQSGSRLKTPSGKYIRIGYADKNEHPYVSI GRYMADKGYL					
g919	DGKAPILGYAEDPVELFFMHIIQSGSRLKTPSGKYIRIGYADKNEHPYVSI GRYMADKGYL					
	250	260	270	280	290	300
	310	320	330	340	350	360
m919.pep	KLQQTSMQGIKSYMRQNPQRLAEVLGQNPSYIFFRELAGSSNDGPVGLGTPLMGEYAGA					
	: : :					
g919	KLQQTSMQGIKAYMRQNPQRLAEVLGQNPSYIFFRELAGSGNEGPGVGLGTPLMGEYAGA					
	310	320	330	340	350	360
	370	380	390	400	410	420
m919.pep	VDRHYITLGAPLFFVATAHPVTRKALNRLIMAQDTGSAIKGAVRVDYFWGYGDEAGELAGK					
	:					
g919	IDRHYITLGAPLFFVATAHPVTRKALNRLIMAQDTGSAIKGAVRVDYFWGYGDEAGELAGK					
	370	380	390	400	410	420
	430	440				
m919.pep	QKTTGYVWQLLPNGMKPEYRPX					
g919	QKTTGYVWQLLPNGMKPEYRPX					
	430	440				

The following partial DNA sequence was identified in *N.meningitidis* <SEQ ID 38>:

a919.seq

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```

1  ATGAAAAAAT ACCTATTCCG CGCGGCCCTG TCGGCATCG CCGCCGCCAT
51  CCTCGCCGCC TGCCAAAGCA AGAGCATCCA AACCTTTCCG CAACCCGACA
101 CATCCGTCAT CAACGGCCCG GACCGGCCG TCGGCATCCC CGACCCCGCC
151 GGAACGACGG TCGGCGGCGG CGGGGCCGTT TATACCGTTG TGCCGCACCT
201 GTCCCTGCCC CACTGGGCGG CGCAGGATTT CGCCAAAAGC CTGCAATCCT
251 TCCGCCTCGG CTGCGCCAAT TTGAAAAACC GCCAAGGCTG GCAGGATGTG
301 TGCGCCCCAAG CCTTTCAAAC CCCCGTCCAT TCCGTTTCAGG CAAAACAGTT
351 TTTTGAACGC TATTTACGCG CGTGGCAGGT TGCAGGCAAC GGAAGCCTTG
401 CCGGTACGGT TACCGGCTAT TACGAGCCGG TGCTGAAGGG CGACGACAGG
451 CGGACGGCAC AAGCCCGCTT CCCGATTTAC GGTATTCCCG ACGATTTTAT
501 CTCCGTCCCC CTGCCTGCCG GTTTGCGGAG CGGAAAAGCC CTTGTCCGCA
551 TCAGGCAGAC GGGAAAAAAC AGCGGCACAA TCACAATAC CGGCGGCACA
601 CATACCGCCG ACCTCTCCCA ATTCCCCTAT ACTGCGCGCA CAACGGCAAT
651 CAAAGGCAGG TTTGAAGGAA GCCGCTTCCT CCCCTACCAC ACGCGCAACC
701 AAATCAACGG CGGCGCGCTT GACGGCAAAG CCCCGATACT CGGTTACGCC
751 GAAGACCCCG TCGAACTTTT TTTTATGCAC ATCCAAGGCT CGGGCCGCTC
801 GAAAACCCCG TCCGGCAAAT ACATCCGCAT CGGCTATGCC GACAAAAACG
851 AACATCCCTA CGTTTCCATC GGACGCTATA TGGCGGACAA AGGCTACCTC
901 AAGCTCGGGC AGACCTCGAT GCAGGGCATC AAAGCCTATA TGCAGCAAAA
951 CCCGCAACGC CTCGCCGAAG TTTTGGGGCA AAACCCAGC TATATCTTTT
1001 TCCGAGAGCT TACCGGAAGC AGCAATGACG GCCCTGTCGG CGCACTGGGC
1051 ACGCCGCTGA TGGGCGAGTA CGCCGGCGCA GTCGACCGGC ACTACATTAC
1101 CTGGGCGCG CCCTTATTTG TCGCCACCGC CCATCCGGTT ACCCGCAAAG
1151 CCCTCAACCG CCTGATTATG GCGCAGGATA CCGGCAGCGC GATTAAAGGC
1201 GCGGTGCGCG TGGATTATTT TTGGGGATAC GCGACGAAG CCGGCGAACT
1251 TGCCGGCAAA CAGAAAACCA CGGGATATGT CTGGCAGCTT CTGCCAACG
1301 GTATGAAGCC CGAATACCG CCGTAA

```

This corresponds to the amino acid sequence <SEQ ID 39; ORF 919.a>:

```

a919.pep
1  MKKYLFRAL CGIAAAILAA CQSKSIQTFP QPDTSVINGP DRPVGIPDPA
51  GTTVGGGGAV YTVVPHLSLP HWAAQDFAKS LQSFRLGCAN LKNRQGWQDV
101 CAQAFQTPVH SVQAKQFFER YFTPWQVAGN GSLAGTVTGY YEPVLKGDDR
151 RTAARFPIY GIPDDFISVP LPAGLRSGKA LVRIQRTGKN SGTIDNTGGT
201 HTADLSQFPI TARTTAIKGR FEGRFLPYH TRNQINGGAL DGKAPILGYA
251 EDPVELFFMH IQSGRLKTP SGKYIRIGYA DKNEHPYVSI GRYMADKGYL
301 KLGQTSMQGI KAYMQQNQR LAEVLGQNPS YIFFRELTGS SNDGPVGALG
351 TPLMGEYAGA VDRHYITLGA PLFVATAHPV TRKALNRLIM AQDTGSAIKG
401 AVRVDYFWGY GDEAGELAGK QKTGYVWQL LPNGMKPEYR P*

```

m919/a919 ORFs 919 and 919.a showed a 98.6% identity in 441 aa overlap

```

10      20      30      40      50      60
m919.pep  MKKYLFRALYGLIAAAILAACQSKSIQTFPQPDTSVINGPDRPVGIPDPAGTTVGGGGAV
          |||
a919      MKKYLFRALCGIAAAILAACQSKSIQTFPQPDTSVINGPDRPVGIPDPAGTTVGGGGAV
          |||
10      20      30      40      50      60

70      80      90      100     110     120
m919.pep  YTVVPHLSLPHWAAQDFAKSLQSFRLGCANLKNRQGWQDVCAQAFQTPVHSFQAKQFFER
          |||
a919      YTVVPHLSLPHWAAQDFAKSLQSFRLGCANLKNRQGWQDVCAQAFQTPVHSVQAKQFFER
          |||
70      80      90      100     110     120

130     140     150     160     170     180
m919.pep  YFTPWQVAGNGSLAGTVTGYEYEPVLKGGDRRTAARFPIYGIPDDFISVPLPAGLRSGKA
          |||
a919      YFTPWQVAGNGSLAGTVTGYEYEPVLKGGDRRTAARFPIYGIPDDFISVPLPAGLRSGKA
          |||
130     140     150     160     170     180

190     200     210     220     230     240
m919.pep  LVRIQRTGKNSGTIDNTGGTHTADLSRFPIARTTAIKGRFEGSRFLPYHTRNQINGGAL
          |||
          :|||

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a919	LVRIRQTGKNSGTIDNTGGTHTADLSQFPITARTTAIKGRFEGSRFLPYHTRNQINGGAL
	190 200 210 220 230 240
m919.pep	DGKAPILGYAEDPVLEFFMHIQGSGRLLKTPSGKYIRIGYADKNEHPYVSIGRYMADKGYL
	250 260 270 280 290 300
a919	DGKAPILGYAEDPVLEFFMHIQGSGRLLKTPSGKYIRIGYADKNEHPYVSIGRYMADKGYL
	250 260 270 280 290 300
m919.pep	KLGGTSMQGIKSYMQRNPQRLAEVLGQNPSYIFFRELAGSSNDGPGVAGLTPLMGEYAGA
	310 320 330 340 350 360
a919	KLGGTSMQGIKSYMQRNPQRLAEVLGQNPSYIFFRELAGSSNDGPGVAGLTPLMGEYAGA
	310 320 330 340 350 360
m919.pep	VDRHYITLGAPLFVATAHPVTRKALNRLIMAQDTGSAIKGAVRVDYFWGYGDEAGELAGK
	370 380 390 400 410 420
a919	VDRHYITLGAPLFVATAHPVTRKALNRLIMAQDTGSAIKGAVRVDYFWGYGDEAGELAGK
	370 380 390 400 410 420
m919.pep	QKTTGYVWQLLPNGMKPEYRPX
	430 440
a919	QKTTGYVWQLLPNGMKPEYRPX
	430 440

121 and 121-1

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 40>:

m121.seq

```

1  ATGGAACAC  AGCTTTACAT  CGGCATCATG  TCGGGAACCA  GCATGGACGG
51  GGCGGATGCC  GTACTGATAC  GGATGGACGG  CGGCAAATGG  CTGGGCGCGG
101 AAGGGCACGC  CTTTACCCCC  TACCCCGGCA  GGTACGCCG  CCAATTGCTG
151 GATTTGCAGG  ACACAGGCGC  AGACGAACTG  CACCGCAGCA  GGATTTTGTC
201 GCAAGAACTC  AGCCGCCTAT  ATGCGCAAAC  CGCCGCCGAA  CTGCTGTGCA
251 GTCAAAACCT  CGCACCGTCC  GACATTACCG  CCCTCGGCTG  CCACGGGCAA
301 ACCGTCCGAC  ACGCGCCGGA  ACACGGTTAC  AGCATACAGC  TTGCCGATTT
351 GCCGCTGCTG  GCGxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx
401 xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx
451 xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx
501 xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx
551 xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx  xxxxxxxxxxx
601 xxxxxxCAGC  TTCCTTACGA  CAAAAACGGT  GCAAAGTCGG  CACAAGGCAA
651 CATATTGCCG  CAACTGCTCG  ACAGGCTGCT  CGCCACCCG  TATTTGCGAC
701 AACGCCACCC  TAAAGACAG  GGGCGCGAAC  TGTTCGCAT  AAATTGGCTC
751 GAAACCTACC  TTGACGGCGG  CGAAAACCGA  TACGACGTAT  TGCGGACGCT
801 TTCCCGTTT  ACCGCGCAA  CCGTTTGCGA  CGCCGTCTCA  CACGCGCGG
851 CAGATGCCCG  TCAAATGTAC  ATTTGCGACG  GCGGCATCCG  CAATCCTGTT
901 TTAATGGCGG  ATTTGGCAGA  ATGTTTCGGC  ACACGCGTTT  CCCTGCACAG
951 CACCGCCGAC  CTGAACCTCG  ATCCGCAATG  GGTGGAAGCC  GCCGnATTTG
1001 CGTGTTTGGC  GCGGTGTTGG  ATTAATCGCA  TTCCCGGTAG  TCCGCACAAA
1051 GCAACCGGCG  CATCAAACC  GTGTATTCTG  AnCGCGGGAT  ATTATTATTG
1101 A

```

This corresponds to the amino acid sequence <SEQ ID 41; ORF 121>:

m121.pep

```

1  METQLYIGIM  SGTSMGADA  VLIRMDGGKW  LGAEGHAFTP  YPGRLRRQLL
51  DLQDTGADEL  HRSRILSQEL  SRLYAQTAAE  LLCSQNLAPS  DITALGCHGQ

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```

101 TVRHAPEHGY SIQLADLPLL Axxxxxxxxx xxxxxxxxxxx xxxxxxxxxxx
151 xxxxxxxxxxx xxxxxxxxxxx xxxxxxxxxxx xxxxxxxxxxx xxxxxxxxxxx
201 xxQLPYDKNG AKSAQGNILP QLLDRLLAHP YFAQRHPKST GRELFAINWL
251 ETYLDGGENR YDVLRTLRSF TAQTVCDAYS HAAADARQMY ICDGGIRNPV
301 LMADLAECFG TRVSLHSTAD LNLDPQWVEA AXFAWLAACW INRIPGSPHK
351 ATGASKPCIL XAGYYY*

```

The following partial DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 42>:

g121.seq

```

1 ATGGAAACAC AGCTTTACAT CGGCATTATG TCGGGAACCA GTATGGACGG
51 GGCGGATGCC GTGCTGGTAC GGATGGACGG CGGCAATATG CTGGGCGCGG
101 AAGGGCACGC CTTTACCCCC TACCCTGACC GGTTCGCGCG CAAATGCTG
151 GATTTGCAGG ACACAGGCAC AGACGAACTG CACCGCAGCA GGATGTTGTC
201 GCAAGAACTC AGCCGCTGT ACAGCGCAAAC CGCCGCCGAA CTGCTGTGCA
251 GTCAAAACCT CGTCCGTGC GACATTACCG CCCTCGGCTG CCACGGGCAA
301 ACCGTCCGAC ACAGCGCCGA ACACGGTtac AGCATACAGC TTGCCGATTT
351 GCCGCTGCTG GCGGAACtGa cgcggatttt TACCGTCggc gacttcCGCA
401 GCCGCGACCT TGCTGCCGGC GGacaAGGTG CGCCGCTCGT CCCCGCCTTT
451 CACGAAGCCC TGTTCCGCGA TGACAGGGAA ACACGCGTGG TACTGAACAT
501 CGGCGGGATT GCCAACATCA GCGTACTCCC CCGCGGCGCA CCGCCTTCG
551 GCTTCGACAC AGGGCCGGGC AATATGCTGA TGGAcgcgtg gacgcaggca
601 cacTGGcagc TGCCTTACGA CAAAacggt gcAAAGcggg cacAAGGCAA
651 catatTGCcg cAACTGCTCG gcaggctGCT CGCCcaccCG TATTTCTCAC
701 AACCccaccc aaAAAGCACG GGgcGCGaac TgtttgcccT AAattggctc
751 gaaacctAcc ttgacggcgg cgaaaaccga tacgacgtat tgcggacgct
801 ttccccgattc accgcgcgcaa ccgTttggga cgccgtctca CACGCAGCGG
851 CAGATGCCCG TCAAATGTAC ATTTGCGGCG GCGGCATCCG CAATCCTGTT
901 TTAATGGCGG ATTTGGCAGA ATGTTTCGGC ACACGCGTTC CCCTGCACAG
951 CACCGCCGAA CTGAACCTCG ATCCTCAATG GGTGGAGGCG gccgCATtTg
1001 cgtggttgGc GGCGTGTtGG ATTAACCGCA TTCCCGGTAG TCCGCACAAA
1051 GCGACCGGCG CATCCAAACC GTGTATTCTG GGCGCGGGAT ATTATTATTG
1101 A

```

This corresponds to the amino acid sequence <SEQ ID 43; ORF 121.ng>:

g121.pep

```

1 METQLYIGIM SGTSMGDADA VLVRMDGGKW LGAEGHAFTP YPDLRLRRKLL
51 DLQDTGTDEL HRSRMLSQEL SRLYAQTAAE LLCSQNLAPC DITALGCHGQ
101 TVRHAPEHGY SIQLADLPLL AELTRIFTVG DFRSRDLAAG GQGAPLVPFAF
151 HEALFRDDRE TRVVLNIGGI ANISVLPPGA PAFGFDTGPG NMLMDAWTQA
201 HWQLPYDKNG AKAAQGNILP QLLGRLLAHP YFSQPHPKST GRELFALNWL
251 ETYLDGGENR YDVLRTLRSF TAQTVWDAYS HAAADARQMY ICGGIRNPV
301 LMADLAECFG TRVSLHSTAE LNLDPQWVEA AAFAWLAACW INRIPGSPHK
351 ATGASKPCIL GAGYYY*

```

ORF 121 shows 73.5% identity over a 366 aa overlap with a predicted ORF (ORF121.ng) from *N. gonorrhoeae*:

m121/g121

	10	20	30	40	50	60
m121.pep	METQLYIGIMSGTSMGDADAVLIRMDGGKWLGAEGHAFTPYPGRLRRQLLDLQDTGADEL					
	: : : : : :					
g121	METQLYIGIMSGTSMGDADAVLVRMDGGKWLGAEGHAFTPYPDLRLRRKLLDLQDTGTDEL					
	10	20	30	40	50	60
	70	80	90	100	110	120
m121.pep	HRSRILSQELSRSLYAQTAAELLCSQNLAPSDITALGCHGQTVRHAPEHGYSIQLADLPLL					
	: : : : : :					
g121	HRSRMLSQELSRSLYAQTAAELLCSQNLAPCDITALGCHGQTVRHAPEHGYSIQLADLPLL					
	70	80	90	100	110	120
	130	140	150	160	170	180

```

al21.seq
1  ATGGAACAC  AGCTTTACAT  CGGCATCATG  TCGGGAACCA  GCATGGACGG
51  GGGCGATGCC  GTACTGATAC  GGATGGACGG  CGGCAAATGG  CTGGGCGCGG
101 AAGGGACGC  CTTTACCCCC  TACCCCGGGA  GGTTACGGCG  CAAATTGCTG
151 GATTTCAGG  ACACAGGCGC  GGACGAAGTG  CACCGCAGCA  GGATGTTGTC
201 GCAAGAACTC  AGCCGCCTGT  ACGCGCAAAC  CGCCGCCGAA  CTGCTGTGCA
251 GTCAAACCT  CGCGCCGTCC  GACATTACCG  CCCTCGGCTG  CCACGGGCAA
301 ACCGTCAGAC  ACGCGCCGGA  ACACAGTTAC  AGCGTACAG  TTGCCGATT
351 GCCGCTGCTG  CGGGAACGGA  CTCAGATTTT  TACCGTCCGG  GACTTCGCGA
401 GCCGCGACCT  TGCGGCCGGC  GGACAAGGCG  CGCCGCTCGT  CCCCGCCTTT
451 CACGAAGCCC  TGTTCCGCGA  CGACAGGGAA  ACACGCGCGG  TACTGAACAT
501 CGGCGGGATT  GCCAACATCA  GCGTACTCCC  CCCCACGCGA  CCCGCCCTCG
551 GCTTCGACAC  AGGACCGGGC  AATATGCTGA  TGGACGCGTG  GATGCAGGCA
601 CACTGGCAGC  TTCTTTACGA  CAAAACCGTG  CGAAAGCGGG  CACAAGGCAA
651 CATATTGCCG  CAACTGCTCG  ACAGGCTGCT  CGCCACCCG  TATTTCGCAC
701 AACCCACCC  TAAAAGCACG  GGGCGCGAAC  TGTTTGCCCT  AAATTGGCTC
751 GAAACCTACC  TTGACGGCGG  CGAAAACCGA  TACGACGTAT  TCGGAGCGCT
801 TTCCCAGATT  ACCGCGCAAA  CCGTTTTCGA  CGCCGTCTCA  CACGCAAGCG
851 CAGATGCCCG  TCAAATGTAC  ATTTGCGGCG  CGGCATCCG  CAATCCTGTT
901 TTAATGGCGG  ATTTGGCAGA  ATGTTTCGGC  ACACGCGTTT  CCCTGCACAG
951 CACCGCCGAA  CTGAACCTCG  ATCCGCAATG  GGTAGAAGCC  GCCGCGTTCG
1001 CATGATGGC  GGC GTGTTGG  GTCAACCGCA  TTCCCGGTAG  TCCGCACAAA
1051 GCAACCGGCG  CATCAAACC  GTGTATTCTG  GGC GCGGGAT  ATTATTATTG
1101 A

```

```

a121.pep
  1  METQLYIGIM  SGTSMDGADA  VLIRMDGGKW  LGAEGHAFTP  YPGRLLRRKLL
    51  DLQDTGADEL  HRSRMLSQEL  SRLYAQTAAE  LLCSQNLAFT  DITALGCHGQ
   101  TVRHAFPEHSY  SVQLADLPLL  AERTQIFTVG  FLFRSDLAAG  GQGAPLVPFA
   151  HEALFRDDRE  TRAVLNIGGI  ANISVLPPDA  PAFGFDTPGP  NMLMDAWMQA
   201  HWQLPYDKNG  AKAAQGNILP  QLLDRLLAHP  YFAQPHPKST  GRELFALNWL
   251  ETYLDGGENR  YDVLRLTSRF  TQQTVFDAVS  HAAADARQMY  ICGGGIRNPV
   301  LMADEAECFG  TRVSLHSTAE  LAQDPQWVEA  AAFAWMAACW  VNRI PGSPHK

```

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351 ATGASKPCIL GAGYYY*

m121/a121 ORFs 121 and 121.a 74.0% identity in 366 aa overlap

m121.pep	10	20	30	40	50	60
	METQLYIGIMSGTSMGADAVLIRMDGGKWLGAEGHAFTYPYGRRLRRQLLDLQDTGADEL					
a121	METQLYIGIMSGTSMGADAVLIRMDGGKWLGAEGHAFTYPYGRRLRRKLLDLQDTGADEL					
	10	20	30	40	50	60
m121.pep	70	80	90	100	110	120
	HRSRILSQELSRLYAQTAELLCSQNLAPSDITALGCHGQTVRHAPHEHGYISQLADLPLL					
a121	HRSRMLSQELSRLYAQTAELLCSQNLAPSDITALGCHGQTVRHAPHSYSVQLADLPLL					
	70	80	90	100	110	120
m121.pep	130	140	150	160	170	180
	XX					
a121	AERTQIFTVGDFRSRDLAAGGQGAFLVPAFHEALFRDDRETRAVLNIGGIANISVLPPDA					
	130	140	150	160	170	180
m121.pep	190	200	210	220	230	240
	XXXXXXXXXXXXXXXXXXXXXXXXXQLPYDKNGAKSAQGNILPQLLDRLLAHPYFAQRHPKST					
a121	PAFGFDTGPGNMLMDAWMQAHWQLPYDKNGAKAAQGNILPQLLDRLLAHPYFAQPHPKST					
	190	200	210	220	230	240
m121.pep	250	260	270	280	290	300
	GRELFAINWLETYLDGGENRYDVLRTLSRFTAQTVCDVASHAAADARQMYICDGGIRNPV					
a121	GRELFAINWLETYLDGGENRYDVLRTLSRFTAQTVFVASHAAADARQMYICGGGIRNPV					
	250	260	270	280	290	300
m121.pep	310	320	330	340	350	360
	LMADLAECFGTRVSLHSTADLNLDPOWVEAAXFAWLAACWINRIPGSPHKATGASKPCIL					
a121	LMADLAECFGTRVSLHSTAELNLDPOWVEAAAFWMAACWVNRIIPGSPHKATGASKPCIL					
	310	320	330	340	350	360
m121.pep	XAGYYYY					
a121	GAGYYYY					

Further work revealed the DNA sequence identified in *N. meningitidis* <SEQ ID 46>:

m121-1.seq

1	ATGGAACAC	AGCTTTACAT	CGGCATCATG	TCGGGAACCA	GCATGGACGG
51	GGCGGATGCC	GTAAGTATAC	GGATGGACGG	CGGCAAATGG	CTGGGCGCGG
101	AAGGGCACGC	CTTTACCCCG	TACCCCGGCA	GGTTACGCCG	CCAATTGCTG
151	GATTTGCAGG	ACACAGGCGC	AGACGAACTG	CACCGCAGCA	GGATTTTGTC
201	GCAAGAACTC	AGCCGCCTAT	ATGCGCAAAC	CGCCGCCGAA	CTGCTGTGCA
251	GTCAAAACCT	CGCACCGTCC	GACATTACCG	CCCTCGGCTG	CCACGGGCAA
301	ACCGTCCGAC	ACGCGCCGGA	ACACGGTTAC	AGCATACAGC	TTGCCGATTT
351	GCCGCTGCTG	GCGGAACGGA	CGCGGATTTT	TACCGTCGGC	GACTTCCGCA
401	GCCGCGACCT	TGCGGCCGGC	GGACAAGGCG	CGCCACTCGT	CCCCGCCTTT
451	CACGAAGCCC	TGTTCCGCGA	CAACAGGGAA	ACACGCGCGG	TACTGAACAT
501	CGGCGGGATT	GCCAAACATCA	GCGTACTCCC	CCCCGACGCA	CCCGCCTTCG
551	GCTTCGACAC	AGGGCCGGGC	AATATGCTGA	TGGACGCGTG	GACGCAGGCA
601	CACTGGCAGC	TTCTTACGA	CAAAAACGGT	GCAAAGGCGG	CACAAGGCAA
651	CATATTGCCG	CAACTGCTCG	ACAGGCTGCT	CGCCACCCG	TATTTGCGAC
701	AACCCACCC	TAAAAGCACG	GGGCGCGAAC	TGTTTGCCCT	AAATTGGCTC
751	GAAACCTACC	TTGACGGCGG	CGAAAACCGA	TACGACGTAT	TGCGGACGCT

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```

801 TTCCCGTTTT ACCGCGCAAA CCGTTTGCGA CGCCGTCTCA CACGCAGCGG
851 CAGATGCCCC TCAAATGTAC ATTTGCGGCG GCGGCATCCG CAATCCTGTT
901 TTAATGGCGG ATTTGGCAGA ATGTTTCGGC ACACGCGTTT CCCTGCACAG
951 CACCGCCGAC CTGAACCTCG ATCCGCAATG GGTGGAAGCC GCCGNATTG
1001 CGTGGTTGGC GCGGTGTTGG ATTAATCGCA TTCCCGGTAG TCCGCACAAA
1051 GCAACCGGCG CATCCAAACC GTGTATTCTG ANCGCGGGAT ATTATTATTG
1101 A

```

This corresponds to the amino acid sequence <SEQ ID 47; ORF 121-1>:

```

m121-1.pep
  1 METQLYIGIM SGTSMGDADA VLIRMDGGKW LGAEGHAFTP YPGLRRQLL
 51 DLQDTGADEL HRSRILSQEL SRLYAQTAAE LLCSQNLAPS DITALGCHGQ
101 TVRHAPHEGY SIQLADLPLL AERTRIFTVG DFRSRDLAAG GQGAPLVPAF
151 HEALFRDNRE TRAVLNIGGI ANISVLPPDA PAFGFDTGPG NMLMDAWTQA
201 HWQLPYDKNG AKAAQGNILP QLLDRLLAHP YFAQPHPKST GRELFALNWL
251 ETYLDGGENR YDVLRTLRSF TAQTVCDAYS HAAADARQMY ICGGGIRNPV
301 LMADLAECFG TRVSLHSTAD LNLDPQWVEA AXFAWLAACW INRIPGSPHK
351 ATGASKPCIL XAGYYY*

```

m121-1/g121 ORFs 121-1 and 121-1.ng showed a 95.6% identity in 366 aa overlap

m121-1.pep	10	20	30	40	50	60
	METQLYIGIMSGTSMGDADAVLIRMDGGKWLGAEHGAFTYPYGLRRQLLDLQDTGADEL					
g121	METQLYIGIMSGTSMGDADAVLVRMDGGKWLGAEHGAFTYPYDRLRRKLLDLQDTGTDEL					
	10	20	30	40	50	60
m121-1.pep	70	80	90	100	110	120
	HRSRILSQELSRLYAQTAAELLCSQNLAPSDITALGCHGQTVRHAPHEGYSIQLADLPLL					
g121	HRSRMLSQELSRLYAQTAAELLCSQNLAPCDITALGCHGQTVRHAPHEGYSIQLADLPLL					
	70	80	90	100	110	120
m121-1.pep	130	140	150	160	170	180
	AERTRIFTVGDFRSRDLAAGGQGAPLVPAFHEALFRDNRETRAVLNIGGIANISVLPPDA					
g121	AELTRIFTVGDFRSRDLAAGGQGAPLVPAFHEALFRDDRETRVVLNIGGIANISVLPPGA					
	130	140	150	160	170	180
m121-1.pep	190	200	210	220	230	240
	PAFGFDTGPGNMLMDAWTQAHWQLPYDKNGAKAAQGNILPQLLDRLLAHPYFAQPHPKST					
g121	PAFGFDTGPGNMLMDAWTQAHWQLPYDKNGAKAAQGNILPQLLGRLLAHPYFSQPHPKST					
	190	200	210	220	230	240
m121-1.pep	250	260	270	280	290	300
	GRELFALNWLETYLDGGENRYDVLRTLRSRFTAQTVCDAYSHAAADARQMYICGGGIRNPV					
g121	GRELFALNWLETYLDGGENRYDVLRTLRSRFTAQTVWDAYSHAAADARQMYICGGGIRNPV					
	250	260	270	280	290	300
m121-1.pep	310	320	330	340	350	360
	LMADLAECFGTRVSLHSTADLNLDPQWVEAAXFAWLAACWINRIPGSPHKATGASKPCIL					
g121	LMADLAECFGTRVSLHSTAELNLDPQWVEAAAFWLAACWINRIPGSPHKATGASKPCIL					
	310	320	330	340	350	360
m121-1.pep	XAGYYYYX					
g121	GAGYYYYX					

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The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 48>:

```
a121-1.seq
1   ATGGAACAC  AGCTTTACAT  CGGCATCATG  TCGGGAACCA  GCATGGACGG
51  GGCGGATGCC  GTACTGATAC  GGATGGACGG  CGGCAAATGG  CTGGGCGCGG
101 AAGGGCACGC  CTTTACCCCC  TACCCGGCA  GGTTACGCCG  CAAATTGCTG
151 GATTTGCAGG  ACACAGGCGC  GGACGAACTG  CACCGCAGCA  GGATGTTGTC
201 GCAAGAACTC  AGCCGCCTGT  ACGCGCAAAC  CGCCGCCGAA  CTGCTGTGCA
251 GTCAAAACCT  CGCGCCGTCC  GACATTACCG  CCCTCGGCTG  CCACGGGCAA
301 ACCGTCAGAC  ACGCGCCGGA  ACACAGTTAC  AGCGTACAGC  TTGCCGATTT
351 GCCGCTGCTG  GCGGAACGGA  CTCAGATTTT  TACCGTCGGC  GACTTCCGCA
401 GCCGCGACCT  TCGCGCCGGC  GGACAAGGCG  CGCCGCTCGT  CCCCGCCTTT
451 CACGAAGCCC  TGTTCCGCGA  CGACAGGGAA  ACACGCGCGG  TACTGAACAT
501 CGGCGGGATT  GCCAACATCA  GCGTACTCCC  CCCCACGCA  CCGCCTTCG
551 GCTTCGACAC  AGGACCGGGC  AATATGCTGA  TGGACGCGTG  GATGCAGGCA
601 CACTGGCAGC  TTCCTTACGA  CAAAACGGT  GCAAAGGCGG  CACAAGGCAA
651 CATATTGCCG  CAACTGCTCG  ACAGGCTGCT  CGCCCACCCG  TATTTGCGAC
701 AACCCACCC  TAAAAGCACG  GGGCGCGAAC  TGTTTGCCCT  AAATTGGCTC
751 GAAACCTACC  TTGACGGCGG  CGAAAACCGA  TACGACGTAT  TGCGGACGCT
801 TTCCCGATT  ACCGCGCAAA  CCGTTTTCGA  CGCCGTCTCA  CACGCAGCGG
851 CACTGCCCCG  TCAAATGTAC  ATTTGCGGCG  GCGGCATCCG  CAATCCTGTT
901 TTAATGGCGG  ATTTGGCAGA  ATGTTTCGGC  ACACGCGTTT  CCCTGCACAG
951 CACCGCCGAA  CTGAACCTCG  ATCCGCAATG  GGTAAGGCC  GCCGCGTTCG
1001 CATGGATGGC  GGCGTGTTGG  GTCAACCGCA  TTCCCGGTAG  TCCGCACAAA
1051 GCAACCGGCG  CATCCAAACC  GTGTATTCTG  GGC GCGGGAT  ATTATTATTG
1101 A
```

This corresponds to the amino acid sequence <SEQ ID 49; ORF 121-1.a>:

```
a121-1.pep
1   METQLYIGIM  SGTSMGADA  VLIRMDGGKW  LGAEGHAFTP  YPGRLLRRKLL
51  DLQDTGADEL  HRSRMLSQEL  SRLYAQTAAE  LLCSQNLAPS  DITALGCHGQ
101 TVRHAPESY  SVQLADLPLL  AERTQIFTVG  DFRSRDLAAG  GQGAPLVPAF
151 HEALFRDDRE  TRAVLNIGGI  ANISVLPPDA  PAFGFDTGPG  NMLMDAWMQA
201 HWQLPYDKNG  AKAAQGNILP  QLLDRLLAHP  YFAQPHPKST  GRELFALNWL
251 ETYLDGGENR  YDVLRTLRF  TAQTVFDAVS  HAAADARQMY  ICGGGIRNPV
301 LMADLAECFG  TRVSLHSTAE  LNLDPQWVEA  AFAWMAACW  VNRIPGSPHK
351 ATGASKPCIL  GAGYYY*
```

m121-1/a121-1 ORFs 121-1 and 121-1.a showed a 96.4% identity in 366 aa overlap

	10	20	30	40	50	60
m121-1.pep	METQLYIGIMSGTSMGADAVLIRMDGGKWLGAEGHAFTPYPGRLRRQLLDLQDTGADEL					
a121-1	METQLYIGIMSGTSMGADAVLIRMDGGKWLGAEGHAFTPYPGRLRRKLLDLQDTGADEL					
	10	20	30	40	50	60
	70	80	90	100	110	120
m121-1.pep	HRSRILSQELSRLYAQTAAELLCSQNLAPSDITALGCHGQTVRHAPHEHGYSIQLADLPLL					
a121-1	HRSRMLSQELSRLYAQTAAELLCSQNLAPSDITALGCHGQTVRHAPESYSVQLADLPLL					
	70	80	90	100	110	120
	130	140	150	160	170	180
m121-1.pep	AERTRIFTVGDFRSRDLAAGGQAPLVPAFHEALFRDNRETRAVLNIGGIANISVLPPDA					
a121-1	AERTQIFTVGDFRSRDLAAGGQAPLVPAFHEALFRDDRETRAVLNIGGIANISVLPPDA					
	130	140	150	160	170	180
	190	200	210	220	230	240
m121-1.pep	PAFGFDTGPGNMLMDAWTQAHWQLPYDKNGAKAAQGNILPQLLDRLLAHPYFAQPHPKST					
a121-1	PAFGFDTGPGNMLMDAWMQAHWQLPYDKNGAKAAQGNILPQLLDRLLAHPYFAQPHPKST					
	190	200	210	220	230	240

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	250	260	270	280	290	300
m121-1.pep	GRELFALNWLETYLDGGENRYDVLRLTSLRFTAQTVCDVASHAAADARQMYICGGGIRNPV					
a121-1	GRELFALNWLETYLDGGENRYDVLRLTSLRFTAQTVFDVASHAAADARQMYICGGGIRNPV					
	250	260	270	280	290	300
	310	320	330	340	350	360
m121-1.pep	LMADLAECFGTRVSLHSTADLNLDLPQWVEAAXFAWLAACWINRIPGSPHKATGASKPCIL					
a121	LMADLAECFGTRVSLHSTAE LNLDLPQWVEAAAFWMAACWVNRI PGSPHKATGASKPCIL					
	310	320	330	340	350	360
m121-1.pep	XAGYYYYX					
a121	GAGYYYYX					

128 and 128-1

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 50>:

```

m128.seq (partial)
1  ATGACTGACA ACGCACTGCT CCATTTGGGC GAAGAACCCC GTTTTGATCA
51  AATCAAAACC GAAGACATCA AACCCGCCCT GCAAACCGCC ATCGCCGAAG
101 CGCGCGAACA AATCGCCGCC ATCAAAGCCC AAACGCACAC CGGCTGGGCA
151 AACACTGTCTG AACCCTGAC CGGCATCACC GAACGCGTCG GCAGGATTTG
201 GGGCGTGGTG TCGCACCTCA ACTGCGTCGC CGACACGCCC GAACTGCGCG
251 CCGTCTATAA CGAACTGATG CCCGAAATCA CCGTCTTCTT CACCGAAATC
301 GGACAAGACA TCGAGCTGTA CAACCGCTTC AAAACCATCA AAAATTCCCC
351 CGAATTCGAC ACCCTCTCCC CCGCACAAAA AACCAAACTC AACCAC
1  TACGCCAGCG AAAAAGTGGC CGAAGCCAAA TACGCGTTCA GCGAAACCGA
51  wGTCAAAAAA TAYTTCCCYG TCGGCAAwGT ATTAACCGGA CTGTTTCGCC
101 AAmTCAAAAAA ACTmTACGGC ATCGGATTTA CCGAAAAAAC yGTCCCCGTC
151 TGGCACAAAG ACGTGCCTTA TTKTGAATTG CAACAAAACG GCGAAmCCAT
201 AGGCGGCGTT TATATGGATT TGTACGCACG CGAAGGCAAA CGCGGCGGCG
251 CGTGGATGAA CGACTACAAA GGCCGCGGCC GTTTTTCAGA CGGCACGCTG
301 CAAYTGCCCA CCGCCTACCT CGTCTGCAAC TTCGCCCCAC CCGTCGCGCG
351 CAGGGAAGCC CGCYTGAGCC ACGACGAAAT CCTCATCCTC TTCCACGAAA
401 CCGGACACGG GCTGCACCAC CTGCTTACCC AAGTGGACGA ACTGGGCGTA
451 TCCGGCATCA ACGGCGTAKA ATGGGACGCG GTCGAACTGC CCAGCCAGTT
501 TATGGAAAAA TTCGTTTGGG AATACAATGT CTTGGCACA mTGTcAGCCC
551 ACGAAGAAAC CGGcgTTCCC yTGCCGAAAG AACTCTTsGA CAAAwTGCTC
601 GCCGCCAAAA ACTTCCAAsG CGGCATGTTC yTsGTCCGGC AAwTGGAGTT
651 CGCCCTCTTT GATATGATGA TTTACAGCGA AGACGACGAA GGCCGTCTGA
701 AAAACTGGCA ACAGGTTTTA GACAGCGTGC GCAAAAAAGT CGCCGTCATC
751 CAGCCGCCCG AATACAACCG CTTCGCCTTG AGCTTCGGCC ACATCTTCGC
801 AGGCGGCTAT TCCGCAGCTn ATTACAGCTA CGCGTGGGCG GAAGTATTGA
851 GCGCGGACGC ATACGCCGCC TTTGAAGAAA GCGACGATGT CGCCGCCACA
901 GGCAAACGCT TTTGGCAGGA AATCCTCGCC GTCGGGGnAT CGCGCAGCGG
951 nGCAGAATCC TTCAAAGCCT TCCGCGGCCG CGAACCGAGC ATAGACGCAC
1001 TCTTGCGCCA CAGCGGTTTC GACAACGCGG TCTGA

```

This corresponds to the amino acid sequence <SEQ ID 51; ORF 128>:

```

m128.pep (partial)
1  MTDNALLHLG EEPRFDQIKT EDIKPALQTA IAEAREQIAA IKAQTHTGWA
51  NTVEPLTGIT ERVGRIWGVV SHLNCVADTP ELRAVYNELM PEITVFFTEI
101 GQDIELYNRF KTIKNSPEFD TLSPAQKTKL NH
//
1  YASEKLREAK YAFSETXVKK YFPVGXVLNG LFAQXKKLYG IGFTEKTVPV

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```

51  WHKDVRYXEL QQNGEXIGGV YMDLYAREGK RGGAWMNDYK GRRRFSDGTL
101 QLPTAYLVCN FAPPVGGREA RLSHDEILIL FHETGHGLHH LLTQVDELGV
151 SGINGVXWDA VELPSQFMEN FVWEYNVLAQ XSAHEETGVP LPKELXDKXL
201 AAKNFQXGMF XVRQXEFALF DMMIYSEDDE GRLKNWQQVL DSVRKKVAVI
251 QPPEYNRFAL SFGHIFAGGY SAAXYSYAWA EVLSADAYAA FEESDDVAAT
301 GKRFWQEILA VGXSRSGAES FKAFRGREPS IDALLRHSGF DNAV*

```

The following partial DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 52>:

```

g128.seq
1  atgattgaca acgCActgct ccacttgggc gaagaaccCC GTTTTaatca
51  aatccaaacc gaagACatca AACCCGCCGT CCAAACCGCC ATCGCCGAAG
101 CGCGCGGACA AATCGCCGCC GTCAAAGCGC AAACGCACAC CGGCTGGGCG
151 AACACCGTCG AGCGTCTGAC CGGCATCACC GAACGCGTCG GCAGGATTGT
201 GGGCGTCGTG TCCCATCTCA ACTCCGTCGT CGACACGCCC GAACGTGGCG
251 CCGTCTATAA CGAACTGATG CCTGAAATCA CCGTCTTCTT CACCGAAATC
301 GGACAAGACA TCGAACTGTA CAACCGCTTC AAAACCATCA AAAATTCCCC
351 CGAATTTGCA ACGCTTTCCC CCGCACAAAA AACCAAGCTC GATCACGACC
401 TGCGCGATTT CGTATTGAGC GGCGCGGAAC TGCCGCCCGA ACGGCAGGCA
451 GCAATGGCAA AACTGCAAAC CGAAGGCGCG CAACTTTCGG CCAAATTCTC
501 CCAAAACGTC CTAGACGCGA CCGACGCGTT CGGCATTAC TTTGACGATG
551 CCGCACCGCT TGCCGGCATT CCCGAAGACG CGCTCGCCAT GTTTGCCGCC
601 GCCGCGCAAA GCGAAGGCAA AACAGGTTAC AAAATCGGCT TGCAGATTCC
651 GCACTACCTT GCCGTTATCC AATACGCCGG CAACCGCGAA CTGCGCGAAC
701 AAATCTACCG CGCCTACGTT ACCCGTGCCA GCGAACTTTC AAACGACGGC
751 AAATTCGACA ACACCGCCAA CATCGACCGC ACGCTCGAAA ACGCATTGAA
801 AACCGccaaa cTGCTCGGCT TTAAAAATTA CGCCGAATTG TCGCTGGCAA
851 CCAAAATGGC GGACACGCCC GAACAGGTTT TAAACTTCCT GCACGACCTC
901 GCCCGCCGCG CCAAACCCTA CGCCGAAAAA GACCTCGCCG AAGTCAAAGC
951 CTTGCGCCGC GAACACCTCG GTCTCGCCGA CCCGACGCCG TGGGACTTGA
1001 GCTACGCCGG CGAAAAACTG CGCGAAGCCA AATACGCATT CAGCGAAACC
1051 GAAGTCAAAA AATACTTCCC CGTCGGCAAA GTTCTGGCAG GCCTGTTTCG
1101 CCAAATCAAA AAATCTACG GCATCGGATT CGCCGAAAAA ACCGTTCCCC
1151 TCTGGCACA AGACGTGCGC TATTTTGAAT TGCAACAAAA CGGCAAAACC
1201 ATCGGCGGCG TTTATATGGA TTTGTACGCA CGCGAAGGCA AACGCGGCGG
1251 CGCGTGGATG AACGActaca AAGGCCGCCG CCGCTTTGCC GACGgcacGC
1301 TGCAACTGCC CACCGCCTAC CTCGTCTGCA ACTTCGCCCC GCCCGTCGGC
1351 GGCAAAGAAG CGCGTTTAAG CCACGACGAA ATCCTCACCC TCTTCCACGA
1401 AacCGGCCAC GGA CTGCACC ACCTGCTTAC CCAAGTGGAC GAACTGGGCG
1451 TGTCCGGCAT CAacggcgtA GAATGGGACG CGGTGGAAC GCCCAGCCAG
1501 TTTATGGAAA ACTTCGTTTG GGAATACAAT GTATTGGCAC AAATGTCCGC
1551 CCACGAAGAA AccgGCGAGC CCCTGCCGAA AGAACTCTTC GACAAAATGC
1601 TcgcCGCCAA AAATTTCCAG CGCGGTATGT TCCTCGTCCG GCAAATGGAG
1651 TTCGCCCTCT TCGATATGAT GATTTACAGT GAAAGCGACG AATGCCGTCT
1701 GAAAACTGG CAGCAGGTTT TAGACAGCGT GCGCAAAGAA GTcGCCGTCA
1751 TCCAACGCC CGAATACAAC CGCTTCGCCA ACAGCTTCGG CCacatctTC
1801 GCcggcGGCT ATTCCGCAGG CTATTACAGC TACGCATGGG CCGAAGTCCt
1851 cAGCACCGAT GCCTACGCCG CCTTTGAAGA AAGcGACGac gtcGCCGCCA
1901 CAGGCAAACG CTTCTGGCAA GAAAtccttg ccgtcggcgg ctCCCCGAGC
1951 gcgGCGGAAT CCTTCAAAGC CTTCCGCGGA CGCGAACCGA GCATAGACGC
2001 ACTGCTGCGC CAaagcggT TCGACAACGC gGcttgA

```

This corresponds to the amino acid sequence <SEQ ID 53; ORF 128.ng>:

```

g128.pep
1  MIDNALLHLG EEPFRNQIQT EDIKPAVQTA IAEARGQIAA VKAQTHTGWA
51  NTVERLTGIT ERVGRIGVV SHLNSVVDTP ELRAVYNELM PEITVFFTEI
101 GQDIELYNRF KTIKNSPEFA TLSPAQKTKL DHDLRDFVLS GAELPPERQA
151 ELAKLQTEGA QLSAKFSQNV LDATDAFGIY FDDAAPLAGI PEDALAMFAA
201 AAQSEGKTGY KIGLQIPHYL AVIQYAGNRE LREQIYRAYV TRASELSNDG

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251 KFDNTANIDR TLENALKTAK LLGFKNYAEL SLATKMADTP EQVLNFLHDL
301 ARRAKPYAEK DLAEVKAFAR EHLGLADPQP WDLSYAGEKL REAKYAFSET
351 EVKKYFPVGK VLAGLFAQIK KLYGIGFAEK TVPVWHKDVR YFELQQNGKT
401 IGGVYMDLYA REGKRGGAWM NDYKGRRRFA DGTLLQLPTAY LVCNFAPPVG
451 GKEARLSHDE ILTLFHETGH GLHLLLTQVD ELGVSGINGV EWDVAVELPSQ
501 FMENFVWEYN VLAQMSAHEE TGEPLPKELF DKMLAAKNFQ RGMFLVRQME
551 FALFDMMIYS ESDECRLKNW QQVLDSVRKE VAVIQPPEYN RFANSFGHIF
601 AGGYSAGYYS YAWAEVLSTD AYAAFEESDD VAATGKRFWQ EILAVGGSRS
651 AAESFKAFRG REPSIDALLR QSGFDNAA*

```

ORF 128 shows 91.7% identity over a 475 aa overlap with a predicted ORF (ORF 128.ng) from *N. gonorrhoeae*:

m128/g128

```

      10      20      30      40      50      60
g128.pep  MIDNALLHLGEEPRFNQIQTEDIKPAVQTAIAEARGQIAAVKAQHTGTWANTVERLTGIT
| | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
m128      MTDNALLHLGEEPRFDQIKTEDIKPALQTAIAEAREQIAAIKAQHTGTWANTVEPLTGIT
      10      20      30      40      50      60

      70      80      90     100     110     120
g128.pep  ERVGRIWGVVSHLNSVVDTPELRAVYNELMPEITVFFTEIGQDIELYNRFKTIKNSPEFA
| | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
m128      ERVGRIWGVVSHLNCVADTPELRAVYNELMPEITVFFTEIGQDIELYNRFKTIKNSPEFD
      70      80      90     100     110     120

      130     140     150     160     170     180
g128.pep  TLSPAQKTKLDHDLRDFVLSGAELPPERQAELAKLQTEGAQLSAKFSQNVLDATDAFGIY
| | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
m128      TLSPAQKTKLNH
      130

//

      340     350     360
g128.pep  YAGEKLREAKYAFSETEVKKYFPVGKVLG
| | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
m128      YASEKLREAKYAFSETXVKYFPVGXVLNG
      10      20      30

      370     380     390     400     410     420
g128.pep  LFAQIKKLYGIGFAEKTVPVWHKDVR YFELQQNGKTIGGVYMDLYAREGKRGGAWMNDYK
| | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
m128      LFAQXKKLYGIGFTEKTVPVWHKDVR YXELQQNGEXIGGVYMDLYAREGKRGGAWMNDYK
      40      50      60      70      80      90

      430     440     450     460     470     480
g128.pep  GRRRFADGTLLQLPTAYLVCNFAPPVVGKEARLSHDEILTLFHETGHGLHLLLTQVDELGV
| | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
m128      GRRRFS DGTLLQLPTAYLVCNFAPPVVGREARLSHDEILILFHETGHGLHLLLTQVDELGV
      100     110     120     130     140     150

      490     500     510     520     530     540
g128.pep  SGINGVEWDAVELPSQFMENFVWEYNVLAQMSAHEETGEPLPKELFDKMLAAKNFQ RGMF
| | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | | |
m128      SGINGVXWDAVELPSQFMENFVWEYNVLAQXSAHEETGVPLPKELXDKMLAAKNFQXGMF
      160     170     180     190     200     210

      550     560     570     580     590     600

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g128.pep      LVRQMEFALFDMMIYSESDCRLKNWQQVLDSVRKEVAVIQPPEYNRFANSFGHIFAGGY
               ||| ||||| ||||| : || ||||| ||||| : ||||| ||||| ||||| |||||
m128          XVRQXEAFALFDMMIYSEDDEGRLKNWQQVLDSVRKKVAVIQPPEYNRFALSFGHIFAGGY
               220       230       240       250       260       270

               610       620       630       640       650       660
g128.pep      SAGYYSYAWAEVLSTDAYAAFEESDDVAATGKRFWQEILAVGGSRSAAESFKAFRGREPS
               ||: ||||| ||||| : ||||| ||||| ||||| ||||| ||||| ||||| |||||
m128          SAAXYSYAWAEVLSADAYAAFEESDDVAATGKRFWQEILAVGXSRSGAESFKAFRGREPS
               280       290       300       310       320       330

               670       679
g128.pep      IDALLRQSGFDNAAX
               ||||| : |||||
m128          IDALLRHSGFDNAVX
               340
```

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al28.seq
1 ATGACTGACA ACGCACTGCT CCATTTGGGC GAAGAACCCC GTTTTGATCA
51 AATCAAAACC GAAGACATCA AACCCGCCCT GCAAACCGCC ATTGCCGAAG
101 CGCGCGAACA AATCGCGGCC ATCAAAGCCC AAACGCAGCT CGGCTGGGCA
151 AACACTGTCTG AACCCTGAC CGGCATCACC GAACGCGTCG GCAGGATTTG
201 GGGCGTGGTG TCGCACCTCA ACTCCGTCAC CGACACGCCC GAACTGCGCG
251 CCGCCTACAA TGAATTAATG CCCGAAATTA CCGTCTTCTT CACCGAAATC
301 GGACAAGACA TCGAGCTGTA CAACCGCTTC AAAACCATCA AAAACTCCCC
351 CGAGTTCGAC ACCCTCTCCC ACGCGCAAAA AACCAAACTC AACCACGATC
401 TGC GCGATTT CGTCTCAGC GCGCGGAAC TGCGGCCCGA ACAGCAGGCA
451 GAATTGGCAA AACTGCAAAC CGAAGGCGCG CAACTTTCCG CCAAATTCTC
501 CCAAACGTC CTAGACGCGA CCGACGCGTT CGGCATTTCAT TTTGACGATG
551 CCGCACCGCT TGCCGGCATT CCCGAAGACG CGCTCGCCAT GTTTGCCGCT
601 GCCGCGCAAA GCGAAGCAA AACAGGCTAC AAAATCGGTT TGCAGATTCC
651 GCACTACCTC GCCGTCATCC AATACGCCGA CAACCGCAA CTGCGCGAAC
701 AAATCTACCG CGCCTAGGTT ACCCGCGCCA GCGAGCTTTC AGACGACGGC
751 AAATTCGACA ACACCGCCA CATCGACCGC ACGCTCGAAA ACGCCCTGCA
801 AACCGCCAAA CTGCTCGGCT TCAAAAATA CGCCGAATTG TCGCTGGCAA
851 CCAAATGGC GGACAGCCCC GAACAAGTTT TAAACTTCCT GCACGACCTC
901 GCCCGCCGCG CCAAACCCTA CGCCGAAAAA GACCTCGCCG AAGTCAAAGC
951 CTTCCGCCGC GAAAGCCTCG GCCTCGCCGA TTTGCAACCG TGGGACTTTG
1001 GCTACGCCCG CGAAAACTG CGCGAAGCCA AATACGCAAT CAGCGAAACC
1051 GAAGTCAAAA AATACTTCCC CGCTCGGCAA GTATTAACAG GACTGTTCCG
1101 CCAAATCAA AAACCTACG GCATCGGATT TACCGAAAAA ACCGTCCCCG
1151 TCTGGCACA AGACGTGCGC TATTTTGAAT TGCAACAAA CGGCGAAACC
1201 ATAGCGGGC TTTTATGGA TTTGTACGCA CGCGAATGGCA AACCGCGCG
1251 CGCGTGGATG AACGACTACA AAGGCGCGCG CCGTTTTCCTA GACGGCAGCG
1301 TGCAACTGCC CACCGCCTAC CTGCTTGCA ACTTCACCCG GCCGTCGCG
1351 GGCAAAGAAG CCCGCTTGAG CCATGACGAA ATCCTCACCC TCTTCCACGA
1401 AACCGGACAC GGCCTGACC ACCTGCTTAC CCAAGTCGAC GAACTGGGCG
1451 TATCCGGCAT CAACGGCGTA GAATGGGACG CAGTCGAAC GCCAGTCAGC
1501 TTTATGAAA AATTCGTTG GGAATACAAT GTCTTGGCGC AAATGTCGCG
1551 CCACGAAGAA ACCGCGTTT CCCTGCCGAA AGAACTCTTC GACAAAATGC
1601 TCGCCGCCAA AAACCTCCAA CGCGGAATGT TCCTCGTCCG CCAAATGGAG
1651 TTCGCCCTCT TTGATATGAT GATTACAGC GAAGACGACG AAGGCCGTCT
1701 GAAAACTGG CAACAGGTTT TAGACAGCGT CGCGAAAGAA GTCGCGCTGT
1751 TCCGACCGCC CGAATACAA CGCTTCGCCA ACAGCTTCGG CCACATCTTC
1801 GCAGGCGGCT ATTCCGCAAG CTATTACAGC TACGCGTGGG CGGAAGTATT
1851 GAGCGCGGAC GCATACGCGC CTTTGAAGA AAGCGACGAT GTCGCGCCA
1901 CAGGCAAACG CTTTTGGCAG GAAATCCTCG CCGTCGCGCG ATCGCGCAGC
1951 GCGGCGAAT CTTTCAAAG CTTCGCGCGA CGCGAACCAG GCATAGACGC
2001 ACTCTTGGCG CACAGCGGCT TCGACAACGC GGCTTGA

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This corresponds to the amino acid sequence <SEQ ID 55; ORF 128.a>:

```

a128.pep
  1  MTDNALLHLG EEPRFDQIKT EDIKPALQTA IAEAREQIAA IKAQHTGTWA
 51  NTVEPLTGIT ERVGRIWGVV SHLNSVTDTP ELRAAYNELM PEITVFFTEI
101  GQDIELYNRF KTIKNSPEFD TLSHAQKTKL NHDLRDFVLS GAELPPEQQA
151  ELAKLQTEGA QLSAKFSQNV LDATDAFGIY FDDAAPLAGI PEDALAMFAA
201  AAQSEGKTGY KIGLQIPHYL AVIQYADNRK LREQIYRAYV TRASELSDDG
251  KFDNTANIDR TLENALQTAK LLGFKNYAEL SLATKMADTP EQVLNFLHDL
301  ARRAKPYAEK DLAEVKAFAR ESIGLADLQP WDLGYAGEKL REAKYAFSET
351  EVKKYFPVGK VLNGLFAQIK KLYGIGFTEK TVPVWHKDVR YFELQQNGET
401  IGGVYMDLYA REGKRGGAWM NDYKGRRRFS DGTQLPTAY LVCNFTPPVG
451  GKEARLSHDE ILTLFHETGH GLHLLTQVD ELGVSGINGV EWDARELPSQ
501  FMENFVWEYN VLAQMSAHEE TGVPLPKELF DKMLAAKNFQ RGMFLVRQME
551  FALFDMMIYS EDDEGRLLKNW QQVLDSVRKE VAVVRPEPEYN RFANSFGHIF
601  AGGYSAGYYS YAWAEVLSAD AYAAFEESDD VAATGKRWFQ EILAVGGSRS
651  AAESFKAFRG REPSIDALLR HSGFDNAA*

```

m128/a128 ORFs 128 and 128.a showed a 66.0% identity in 677 aa overlap

```

m128.pep      10      20      30      40      50      60
MTDNALLHLGEEPRFDQIKTEDIKPALQTAIAEAREQIAAIIKAQHTGTWANTVEPLTGIT
a128          10      20      30      40      50      60
MTDNALLHLGEEPRFDQIKTEDIKPALQTAIAEAREQIAAIIKAQHTGTWANTVEPLTGIT

m128.pep      70      80      90     100     110     120
ERVGRIWGVVSHLNCVADTPELRAVYNELMPEITVFFTEIGQDIELYNRFKTIKNSPEFD
a128          70      80      90     100     110     120
ERVGRIWGVVSHLNSVTDTPELRAAYNELMPEITVFFTEIGQDIELYNRFKTIKNSPEFD

m128.pep      130
TLSPAQKTKLNH-----
a128          130     140     150     160     170     180
TLSHAQKTKLNHDLRDFVLSGAELPPEQQAELAKLQTEGAQLSAKFSQNVLDATDAFGIY

m128.pep      -----
a128          190     200     210     220     230     240
FDDAAPLAGIPEDALAMFAAAAQSEGKTGYKIGLQIPHYLAVIQYADNRKLREQIYRAYV

m128.pep      -----
a128          250     260     270     280     290     300
TRASELSDDGKFDNTANIDRTLENALQTAKLLGFKNYAELSLATKMADTPEQVLNFLHDL

m128.pep      -----140     150
-----YASEKLREAKYAFSETXVKKYFPVGX
a128          310     320     330     340     350     360
ARRAKPYAEKDLAEVKAFARESLGLADLQPWDLYAGEKLREAKYAFSETEVKKYFPVGK

m128.pep      160     170     180     190     200     210
VLNGLFAQXKKLYGIGFTEKTVPVWHKDVRXYELQQNGEXIGGVYMDLYAREGKRGGAWM
a128          370     380     390     400     410     420
VLNGLFAQIKKLYGIGFTEKTVPVWHKDVRXYFELQQNGETIGGVYMDLYAREGKRGGAWM

m128.pep      220     230     240     250     260     270
NDYKGRRRFS DGTQLPTAYLVCNFAPPVGGREARLSHDEILILFHETGHGLHLLTQVD

```

a128	NDYKGRRRFS	DGTLQLPTAYLVCN	FTPPVGGKEARLSH	DEILTLFHETGHGL	HLLTQVD	
	430	440	450	460	470	480
m128.pep	280	290	300	310	320	330
	ELGVSGINGV	XWDAVELPSQFMEN	FVWEYNVLAQXSA	HEETGVPLPKELX	DKXLAAKNFQ	
a128	ELGVSGINGV	EWDAVELPSQFMEN	FVWEYNVLAQMSA	HEETGVPLPKELF	DKMLAAKNFQ	
	490	500	510	520	530	540
m128.pep	340	350	360	370	380	390
	XGMFXVRQXE	FALFDMMIYSEDDE	GRLNWQQVLD	SVRKKVAVIQPPE	YNRFALSFGHIF	
a128	RGMFLVRQME	FALFDMMIYSEDDE	GRLNWQQVLD	SVRKEVAVVRPPE	YNRFANSFGHIF	
	550	560	570	580	590	600
m128.pep	400	410	420	430	440	450
	AGGYSAAXY	SYAWAEVLSADAY	AAFEESDDVAATG	KREWQEILAVGXS	RSGAESFKAFRG	
a128	AGGYSAGY	SYAWAEVLSADAY	AAFEESDDVAATG	KREWQEILAVGGS	RSAAESFKAFRG	
	610	620	630	640	650	660
m128.pep	460	470				
	REPSIDALLR	HSGFDNAVX				
a128	REPSIDALLR	HSGFDNAAX				
	670					

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m128-1.seq
1  ATGACTGACA  ACGCACTGCT  CCATTTGGGC  GAAGAACCCC  GTTTTGATCA
51  AATCAAAACC  GAAGACATCA  AACC CGCCCT  GCAAACCGCC  ATCGCCGAAG
101 CGCGCGAACA  AATCGCCGCC  ATCAAAGCCC  AAACGCACAC  CGGTGGGCA
151 AACACTGTCG  AACCCTGAC  CGCATCACC  GAACGCGTCG  CAGGAGTTTG
201 GGGCGTGGTG  TCGCACCTCA  ACTCCGTGCG  CGACACGCCC  GAACTGCGCG
251 CCGTCTATAA  CGAAGTGATG  CCCGAAATCA  CCGTCTTCTT  CACCGAAATC
301 GGACAAGACA  TCGAGCTGTA  CAACCGCTTC  AAAACCATCA  AAAATTCCCC
351 CGAATTTCGAC  ACCCTCTCCC  CGCACAAAA  AACCAAATC  AACCAGATC
401 TGC GCGATTT  CGTCTCAGC  GCGCGGAAAC  TGCCGCCCGA  CACGACGGCA
451 GAACTGGCAA  AACTGCAAAC  CGAAGGCGCG  CAACTTTCCG  CCAAATTCTC
501 CCAAAACGTC  CTAGACGCGA  CCGACGCGTT  CGGCATTTAC  TTTGACGATG
551 CCGCACCCGT  TGCCGGCATT  CCCGAAGACG  CGCTCGCCAT  GTTTGCCCGC
601 CGCGCGCAAA  GCGAAAGCAA  AACAGGCTAC  AAAATCGGT  TGCAGATTCC
651 ACACTACCTC  GCGCTCATCC  AATACGCCGA  CAACCGCGAA  CTGCGCGAAC
701 AAATCTACCG  CGCTACGTT  ACCGCGGCCA  GCGAACTTTC  AGACGACGGC
751 AAATTCGACA  ACACCGCCAA  CATCGACCGC  ACGCTCGCAA  ACGCCCTGCA
801 AACCGCCAAA  CTGCTCGGCT  TCAAAAACTA  CGCCGAATTG  TCGCTGGCAA
851 CCAAAATGGC  GGACACGCC  GAACAAGTTT  TAAACTTCT  CACGACCTC
901 GCCCGCGCGC  CCAAACCTA  CGCGCAAAAA  GACCTCGCCG  AAGTCAAAGC
951 CTTGCCCCGC  GAAAGCCTGA  ACCTCGCCGA  TTTGCAACCG  TGGGACTTGG
1001 GCTACGCCAG  CGAAAACTG  CGCGAAGCCA  AATACGCGTT  CAGCGAAACC
1051 GAAGTCAAAA  AATACTTCCC  CGTCGGCAAA  GTATTAAACG  GACTGTTTCG
1101 CCAAACTAAA  AAACCTACG  GCATCGGATT  TACCGAAAAA  ACCGTCCCCG
1151 TCTGGCACAA  AGACGTGCGC  TATTTTGAAT  TGCAACAAAA  CGCGGAAACC
1201 ATAGGCGGCG  TTTATATGGA  TTTGTACGCA  CGCGAAGGCA  AACGCGGCGG
1251 CGCGTGGATG  AACGACTACA  AAGGCCGCGC  CCGTTTTTCA  GACGGCACGC
1301 TGCAACTGCC  CACCGCCTAC  CTGCTGCA  ACTTCGCCCC  ACCCGTCGGC
1351 GGCAGGGAAG  CCCGCTGAG  CCACGAGAA  ATCTCATCC  TCTCCACGA
1401 AACCGGACAC  GGGCTGCACC  ACCTGCTTAC  CCAAGTGGAC  GAACTGGGCG

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1451 TATCCGGCAT CAACGGCGTA GAATGGGACG CGGTGGAAC T GCCCAGCCAG
1501 TTTATGGAAA ATTTTCGTTTG GGAATACAAT GTCTTGGCAC AAATGTCAGC
1551 CCACGAAGAA ACCGGCGTTC CCCTGCCGAA AGAACTCTTC GACAAAATGC
1601 TCGCCGCCAA AAACCTCCAA CGCGGCATGT TCCTCGTCCG GCAAATGGAG
1651 TTCGCCCTCT TTGATATGAT GATTTACAGC GAAGACGACG AAGGCCGTCT
1701 GAAAAACTGG CAACAGGTTT TAGACAGCGT GCGCAAAAAA GTCGCCGTCA
1751 TCCAGCCGCC CGAATACAAC CGCTTCGCCT TGAGCTTCGG CCACATCTTC
1801 GCAGGCGGCT ATTCGCGCAG CTATTACAGC TACGCGTGGG CGGAAGTATT
1851 GAGCGCGGAC GCATACGCCG CCTTTGAAGA AAGCGACGAT GTCGCCGCCA
1901 CAGGCAAACG CTTTGGCAG GAAATCCTCG CCGTCGGCGG ATCGCGCAGC
1951 GCGGCAGAA CTTCAAAGC CTTCCGCGGC CGCGAACCGA GCATAGACGC
2001 ACTCTTGC GC CACAGCGGTT TCGACAACGC GGTCTGA

```

This corresponds to the amino acid sequence <SEQ ID 57; ORF 128-1>:

m128-1.pep.

```

1 MTDNALLHLG EEPRFDQIKT EDIKPALQTA IAEAREQIAA IKAQTHTGWA
51 NTVEPLTGIT ERVGRWGVV SHLNSVADTP ELRAVYNELM PEITVFTEI
101 GQDIELYNRF KTIKNSPEFD TLSPAQKTKL NHDLRDFVLS GAELPPEQQA
151 ELAKLQTEGA QLSAKFSQNV LDATDAFGIY FDDAAPLAGI PEDALAMFAA
201 AAQSESKTGY KIGLQIPHYL AVIQYADNRE LREQIYRAYV TRASELSDDG
251 KFDNTANIDR TLANALQTAK LLGFKNYAEL SLATKMADTP EQVLNLFHDL
301 ARRAKPYAEK DLAEVKAFAR ESLNLADLQP WDLGYASEKL REAKYAFSET
351 EVKKYFPVGK VLNGLFAQIK KLYGIGFTEK TVPVWHKDVR YFELQQNGET
401 IGGVYMDLYA REGKRGGAWM NDYKGRRRFS DGTQLQLPTAY LVCNFAPPVG
451 GREARLSHDE ILILFHETGH GLHLLLTQVD ELGVSGINGV EWDVAVELPSQ
501 FMENFVWEYN VLAQMSAHEE TGVPLPKELF DKMLAAKNFQ RGMFLVRQME
551 FALFDMMIYS EDDEGRLKNW QQVLDSVRKK VAVIQPPEYN RFALSFGHIF
601 AGGYSAGYYS YAWAEVLSAD AYAAFEESDD VAATGKRFWQ EILAVGGSRS
651 AAESFKAFRG REPSIDALLR HSGFDNAV*

```

The following partial DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 58>:

g128-1.seq (partial)

```

1 ATGATTGACA ACGCACTGCT CCACTTGGGC GAAGAACCCC GTTTTAATCA
51 AATCAAAACC GAAGACATCA AACC CGCGT CCAAACCGCC ATCGCCGAAG
101 CGCGCGGACA AATCGCGCGC GTCAAAGCGC AAACGCACAC CGGCTGGCG
151 AACACCGTCG AGCGTCTGAC CGGCATCACC GAACGCGTCG GCAGGATTTG
201 GGGCGTCGTG TCCCATCTCA ACTCCGTCGT CGACACGCCC GAACTGCGCG
251 CCGTCTATAA CGAACTGATG CCTGAAATCA CCGTCTTCTT CACCGAAATC
301 GGACAAGACA TCGAACTGTA CAACCGCTTC AAAACCATCA AAAATTCCCC
351 CGAATTTGCA ACGCTTTCCC CCGCACAAAA AACCAAGCTC GATCAGACC
401 TGCGCGATTT CGTATTGAGC GGCGCGGAAC TGCCGCCCGA ACGGCAGGCA
451 GAACTGGCAA AACTGCAAC CGAAGGCGCG CAACTTTCCG CCAAATTCTC
501 CCAAACGTC CTAGACGCGA CCGACGCGTT CGGCATTTAC TTTGACGATG
551 CCGCACCGCT TGCCGGCATT CCCGAAGACG CGCTCGCCAT GTTTGCCGCC
601 GCGCGCGCAA GCGAAGGCAA AACAGGTTAC AAAATCGGCT TGCAGATTCC
651 GCAC TACCTT GCCGTTATCC AATACGCCGG CAACCGCGAA CTGCGCGAAC
701 AAATCTACCG CGCCTACGTT ACCCGTGCCA GCGAACTTTC AAACGACGGC
751 AAATTCGACA ACACCGCCAA CATCGACCGC ACGCTCGAAA ACGCATTGAA
801 AACC GCCAAA CTGCTCGGCT TAAAAATTA CGCCGAATTG TCGCTGGCAA
851 CAAAAATGGC GGACACGCCG GAACAGGTTT TAAACTTCCT GCACGACCTC
901 GCGCGCGCG CCAAACCTA CGCCGAAAAA GACCTCGCCG AAGTCAAAGC
951 CTTCGCGCG GAACACCTCG GTCTCGCCGA CCCGACGCCG TGGGACTTGA
1001 GCTACGCCGG CGAAAACTG CGCGAAGCCA AATACGCATT CAGCGAAACC
1051 GAAGTCAAAA AATACTTCCC CGTCGGCAA GTTCTGGCAG GCCTGTTCCG
1101 CCAAATCAAA AAACCTTACG GCATCGGATT CGCCGAAAAA ACCGTTCCCG
1151 TCTGGCACAA AGACGTGCGC TATTTTGAAT TGCAACAAAA CGGCAAAACC
1201 ATCGGCGGCG TTTATATGGA TTTGTACGCA CGCGAAGGCA AACGCGCGCG
1251 CGCGTGGATG AACGACTACA AAGGCCGCCG CCGCTTTGCC GACGCGACGC
1301 TGCAACTGCC CACCGCCTAC CTCGTCTGCA ACTTCGCCCC GCCCGTCGGC
1351 GGC AAAAGAG CGCGTTTAAG CCACGACGAA ATCCTCACCC TCTTCCACGA
1401 AACC GCCCAC GGACTGCACC ACCTGCTTAC CCAAGTGGAC GAACTGGGCG
1451 TGTCCGGCAT CAACGGCGTA AAA

```

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This corresponds to the amino acid sequence <SEQ ID 59; ORF 128-1.ng>:

g128-1.pep (partial)

```

1  MIDNALLHLG EEPRFNQIKT EDIKPAVQTA IAEARGQIAA VKAQHTGTGWA
51  NTVRLTGIT ERVGRIWGVV SHLNSVVDTP ELRAVYNELM PEITVFFTEI
101 GQDIELYNRF KTIKNSPEFA TLSPAQKTKL DHDLRDFVLS GAELPPERQA
151 ELAKLQTEGA QLSAKFSQNV LDATDAFGIY FDDAAPLAGI PEDALAMFAA
201 AAQSEGKTGY KIGLQIPHYL AVIQYAGNRE LREQIYRAYV TRASELSNDG
251 KFDNTANIDR TLENALKTAK LLGFKNYAEL SLATKMADTP EQVLNFLHDL
301 ARRAKPYAEK DLAEVKAFAR EHLGLADPQP WDLSYAGEKL REAKYAFSET
351 EVKKYFPVGK VLAGLFAQIK KLYGIGFAEK TVPVWHKDVR YFELQQNGKT
401 IGGVMDLYA REGKRGGAWM NDYKGRRRFA DGTQLQPTAY LVCNFAPPVG
451 GKEARLSHDE ILTLFHETGH GLHLLTQVD ELGVSINGV K

```

m128-1/g128-1 ORFs 128-1 and 128-1.ng showed a 94.5% identity in 491 aa overlap

g128-1.pep	10	20	30	40	50	60
	MIDNALLHLG	EEPRFNQIKT	EDIKPAVQTA	IAEARGQIAA	VKAQHTGTGWA	NTVRLTGIT
m128-1	MTDNALLHLG	EEPRFDQIKT	EDIKPAVQTA	IAEAREQIAA	IKAQHTGTGWA	NTVRLTGIT
	10	20	30	40	50	60
g128-1.pep	70	80	90	100	110	120
	ERVGRIWGVV	SHLNSVVDTP	PELRAVYNELM	PEITVFFTEI	GQDIELYNRF	KTIKNSPEFA
m128-1	ERVGRIWGVV	SHLNSVADTP	PELRAVYNELM	PEITVFFTEI	GQDIELYNRF	KTIKNSPEFD
	70	80	90	100	110	120
g128-1.pep	130	140	150	160	170	180
	TLSPAQKTKL	DHDLRDFVLS	GAELPPERQA	ELAKLQTEGA	QLSAKFSQNV	LDATDAFGIY
m128-1	TLSPAQKTKL	NHDLRDFVLS	GAELPPERQA	ELAKLQTEGA	QLSAKFSQNV	LDATDAFGIY
	130	140	150	160	170	180
g128-1.pep	190	200	210	220	230	240
	FDDAAPLAGI	PEDALAMFAA	AAQSEGKTGY	KIGLQIPHYL	AVIQYAGNRE	LREQIYRAYV
m128-1	FDDAAPLAGI	PEDALAMFAA	AAQSEKTKGY	KIGLQIPHYL	AVIQYADNRE	LREQIYRAYV
	190	200	210	220	230	240
g128-1.pep	250	260	270	280	290	300
	TRASELSNDG	KFDNTANIDR	TLENALKTAK	LLGFKNYAEL	SLATKMADTP	EQVLNFLHDL
m128-1	TRASELSDDG	KFDNTANIDR	TLANALQTA	LLGFKNYAEL	SLATKMADTP	EQVLNFLHDL
	250	260	270	280	290	300
g128-1.pep	310	320	330	340	350	360
	ARRAKPYAEK	DLAEVKAFAR	EHLGLADPQP	WDLSYAGEKL	REAKYAFSET	EVKKYFPVGK
m128-1	ARRAKPYAEK	DLAEVKAFAR	ESLNADLPQ	WDLGYASEKL	REAKYAFSET	EVKKYFPVGK
	310	320	330	340	350	360
g128-1.pep	370	380	390	400	410	420
	VLAGLFAQIK	KLYGIGFAEK	TPPVWHKDVR	YFELQQNGKT	IGGVMDLYA	REGKRGGAWM
m128-1	VNLGLFAQIK	KLYGIGFTEK	TPPVWHKDVR	YFELQQNGKT	IGGVMDLYA	REGKRGGAWM
	370	380	390	400	410	420
g128-1.pep	430	440	450	460	470	480
	NDYKGRRRFA	DGTQLQPTAY	LVCNFAPPVG	GKEARLSHDE	ILTLFHETGH	GLHLLTQVD

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```

|||||||:|||||||:|||||||:|||||||
m128-1 NDYKGRRRFSDGTLQLPTAYLVCNFAPPVGGREARLSHDEILILFHETGHLHLLTQVD
          430      440      450      460      470      480

          490
g128-1.ppe ELGVSGINGVK
          |||||:
m128-1 ELGVSGINGVEWDAVELPSQFMENFVWEYNVLAQMSAHEETGVPLPKELFDKMLAAKNFQ
          490      500      510      520      530      540

```

The following DNA sequence was identified in *N. meningitidis* <SEQ ID 60>:

```

a128-1.seq
1  ATGACTGACA ACGCACTGCT CCATTTGGGC GAAGAACCCC GTTTTGATCA
51  AATCAAAACC GAAGACATCA AACCCGCCCT GCAAACCGCC ATTGCCGAAG
101 CGCGCGAACA AATCGCCGCC ATCAAAGCCC AAACGCACAC CGGCTGGGCA
151 AACACTGTCTG AACCCCTGAC CGGCATCACC GAACGCGTCG GCAGGATTTG
201 GGGCGTGGTG TCGCACCTCA ACTCCGTCAC CGACACGCCC GAACTGCGCG
251 CCGCCTACAA TGAATTAATG CCCGAAATTA CCGTCTTCTT CACCGAAATC
301 GGACAAGACA TCGAGCTGTA CAACCGCTTC AAAACCATCA AAAACTCCCC
351 CGAGTTTCGAC ACCCTCTCCC ACGCGCAAAA AACCAAACTC AACCACGATC
401 TGCGCGATTT CGTCTCAGC GCGCGGAAC TGCCGCCCGA ACAGCAGGCA
451 GAATTGGCAA AACTGCAAA CGAAGGCGCG CAACTTTCCT CCAAATTCCT
501 CCAAACGTC CTAGACGCGA CCGACGCGTT CGGCATTAC TTTGACGATG
551 CCGCACCGCT TGCCGGCATT CCCGAAGACG CGCTCGCCAT GTTTGCCGCT
601 GCGCGCAAA GCGAAGGCAA AACAGGCTAC AAAATCGGTT TGCAGATTCC
651 GCACTACCTC GCCGTCATCC AATACGCCGA CAACCGCAA CTGCGCGAAC
701 AAATCTACCG CGCCTACGTT ACCCGCGCCA GCGAGCTTTC AGACGACGCG
751 AAATTCGACA ACACCGCCAA CATCGACCGC ACGCTCGAAA ACGCCCTGCA
801 AACCGCCAAA CTGCTCGGCT TCAAAAATA CGCCGAATTG TCGCTGGCAA
851 CCAAATGGC GGACACCCCC GAACAAGTTT TAAACTTCCT GCACGACCTC
901 GCGCGCCGCG CCAAACCCTA CGCCGAAAAA GACCTCGCCG AAGTCAAAGC
951 CTTCGCCCCG GAAAGCCTCG GCCTCGCCGA TTTGCAACCG TGGGACTTGG
1001 GCTACGCCGG CGAAAAACTG CGCGAAGCCA AATACGCATT CAGCGAAACC
1051 GAAGTCAAAA AATACTTCCC CGTCGGCAAA GTATTAAACG GACTGTTGCG
1101 CCAAATCAA AAACCTCTACG GCATCGGATT TACCGAAAAA ACCGTCCCCG
1151 TCTGGCACA AGACGTGCGC TATTTTGAAT TGCAACAAA CGGCGAAACC
1201 ATAGGCGGCG TTTATATGGA TTTGTACGCA CGCGAAGGCA AACGCGGCGG
1251 CGCGTGGATG AACGACTACA AAGGCCGCCG CCGTTTTTCA GACGGCACGC
1301 TGCAACTGCC CACCGCCTAC CTCGTCTGCA ACTTACCCC GCCGTCGCG
1351 GGCAAAGAAG CCCGCTTGAG CCATGACGAA ATCCTCACCC TCTTCCACGA
1401 AACCGGACAC GGCCTGCACC ACCTGCTTAC CCAAGTCGAC GAACTGGGCG
1451 TATCCGGCAT CAACGGCGTA GAATGGGACG CAGTCGAACT GCCCAGTCAG
1501 TTTATGGAAA ATTTCTGTTG GGAATACAAT GTCTTGGCGC AAATGTCCCG
1551 CCACGAAGAA ACCGCGCTTC CCCTGCCGAA AGAACTCTTC GACAAAATGC
1601 TCGCCGCCAA AAACCTCCAA CGCGGAATGT TCCTCGTCCG CCAAATGGAG
1651 TTCGCCCTCT TTGATATGAT GATTTACAGC GAAGACGACG AAGGCCGTCT
1701 GAAAAACTGG CAACAGGTTT TAGACAGCGT GCGCAAAGAA GTCGCCGTCG
1751 TCCGACCGCC CGAATACAAC CGCTTCGCCA ACAGCTTCGG CCACATCTTC
1801 GCAGGCGGCT ATTCCGACG CTATTACAGC TACGCGTGGG CGGAAGTATT
1851 GAGCGCGGAC GCATACGCCG CCTTTGAAGA AAGCGACGAT GTCGCCGCCA
1901 CAGGCAAACG CTTTTGGCAG GAAATCCTCG CCGTCGGCGG ATCGCGCAGC
1951 GCGGCAGAA CTCTCAAAGC CTCCGCGGGA CGCGAACCGA GCATAGACGC
2001 ACTCTTGCGC CACAGCGGCT TCGACAACGC GGCTTGA

```

This corresponds to the amino acid sequence <SEQ ID 61; ORF 128-1.a>:

```

a128-1.ppe
1  MTDNALLHLG EEPRFDQIKT EDIKPALQTA IAEAREQIAA IKAQTHTGWA
51  NTVEPLTGIT ERVGRWGVV SHLNSVTDTP ELRAAYNELM PEITVFFTEI
101 GQDIELYNRF KTIKNSPEFD TLSHAQKTKL NHDLRDFVLS GAELPPEQQA
151 ELAKLQTEGA QLSAKFSQNV LDATDAFGIY FDDAAPLAGI PEDALAMFAA
201 AAQSEGKTY KIGLQIPHYL AVIQYADNRK LREQIYRAYV TRASELSDDG
251 KFDNTANIDR TLENALQTAK LLGFKNYAEI SLATKMADTP EQVLNFLHDL

```

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```

301  ARRAKPYAEK  DLAEVKAFAR  ESLGLADLQP  WDLGYAGEKL  REAKYAFSET
351  EVKKYFPVVK  VLNGLFAQIK  KLYGIGFTEK  TVPVWHKDVR  YFELQQNGET
401  IGGVYMDLYA  REGKRGGAWM  NDYKGRRRFS  DGTQLQPTAY  LVCNFTPPVG
451  GKEARLSHDE  ILTLFHETGH  GLHLLLTQVD  ELGVSGINGV  EWDAVELPSQ
501  FMENFVWEYN  VLAQMSAHEE  TGVPLPKELF  DKMLAAKNFQ  RGMFLVRQME
551  FALFDMMIYS  EDDEGRLKNW  QQVLDSVRKE  VAVVRPEYN  RFANSFGHIF
601  AGGYSAGYYS  YAWAEVLSAD  AYAAFEESDD  VAATGKRFWQ  EILAVGGSRS
651  AAESFKAFRG  REPSIDALLR  HSGFDNAA*

```

m128-1/a128-1 ORFs 128-1 and 128-1.a showed a 97.8% identity in 677 aa overlap

	10	20	30	40	50	60
a128-1.pep	MTDNALLHLGEEPRFDQIKTEDIKPALQTAIAEAREQIAAIKAQHTGTGWANTVEPLTGIT					
m128-1	MTDNALLHLGEEPRFDQIKTEDIKPALQTAIAEAREQIAAIKAQHTGTGWANTVEPLTGIT					
	10	20	30	40	50	60
	70	80	90	100	110	120
a128-1.pep	ERVGRIWGVVSHLNSVTDTPELRAAYNELMPEITVFFTEIGQDIELYNRFKTIKNSPEFD					
m128-1	ERVGRIWGVVSHLNSVADTPELRAVYNELMPEITVFFTEIGQDIELYNRFKTIKNSPEFD					
	70	80	90	100	110	120
	130	140	150	160	170	180
a128-1.pep	TLSHAQKTKLNHDLRDFVLSGAELPPEQQAELAKLQTEGAQLSAKFSQNVLDATDAFGIY					
m128-1	TLSPAQKTKLNHDLRDFVLSGAELPPEQQAELAKLQTEGAQLSAKFSQNVLDATDAFGIY					
	130	140	150	160	170	180
	190	200	210	220	230	240
a128-1.pep	FDDAAPLAGIPEDALAMFAAAAQSEKGTGYKIGLQIPHYLAVIQYADNRKLREQIYRAYV					
m128-1	FDDAAPLAGIPEDALAMFAAAAQSESKTGYKIGLQIPHYLAVIQYADNRELREQIYRAYV					
	190	200	210	220	230	240
	250	260	270	280	290	300
a128-1.pep	TRASELSDDGKFDNTANIDRTLENALQTAKLLGFKNYAELSLATKMADTPEQVLNFLHDL					
m128-1	TRASELSDDGKFDNTANIDRTLANALQTAKLLGFKNYAELSLATKMADTPEQVLNFLHDL					
	250	260	270	280	290	300
	310	320	330	340	350	360
a128-1.pep	ARRAKPYAEKDLAEVKAFARESLGLADLQPWDLYAGEKLREAKYAFSETEVKKYFPVVK					
m128-1	ARRAKPYAEKDLAEVKAFARESLNLADLQPWDLYASEKLREAKYAFSETEVKKYFPVVK					
	310	320	330	340	350	360
	370	380	390	400	410	420
a128-1.pep	VLNGLFAQIKKLYGIGFTEKTVPVWHKDVRVYFELQQNGETIGGVYMDLYAREGKRGGAWM					
m128-1	VLNGLFAQIKKLYGIGFTEKTVPVWHKDVRVYFELQQNGETIGGVYMDLYAREGKRGGAWM					
	370	380	390	400	410	420
	430	440	450	460	470	480
a128-1.pep	NDYKGRRRFSDGTQLQPTAYLVCNFTPPVGGKEARLSHDEILTLFHETGHGLHLLLTQVD					
m128-1	NDYKGRRRFSDGTQLQPTAYLVCNFAPPVGGREARLSHDEILILFHETGHGLHLLLTQVD					
	430	440	450	460	470	480
	490	500	510	520	530	540
a128-1.pep	ELGVSGINGVEWDAVELPSQFMENFVWEYNVLAQMSAHEETGVPLPKELFDKMLAAKNFQ					

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m128-1	ELGVSGINGVEWDAVELPSQFMENFVWEYNVLAQMSAHEETGVPLPKELFDKMLAAKNFQ
	490 500 510 520 530 540
a128-1.pep	RGMFLVRQMEFALFDMMIYSEDDEGRLKNWQQVLDVSRKEVAVVRPPEYNRFANSFGHIF
	550 560 570 580 590 600
m128-1	RGMFLVRQMEFALFDMMIYSEDDEGRLKNWQQVLDVSRKKVAVIQPPEYNRFALSFGHIF
	550 560 570 580 590 600
a128-1.pep	AGGYSAGYYSYAWAEVLSADAYAAFEESDDVAATGKRFWQEILAVGGSRSAESFKAFRG
	610 620 630 640 650 660
m128-1	AGGYSAGYYSYAWAEVLSADAYAAFEESDDVAATGKRFWQEILAVGGSRSAESFKAFRG
	610 620 630 640 650 660
a128-1.pep	REPSIDALLRHSGFDNAAX
	670 679
m128-1	REPSIDALLRHSGFDNAVX
	670

206

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 62>:

```

m206.seq
1  ATGTTTCCCC CCGACAAAAC CCTTTTCCTC TGTCTCAGCG CACTGCTCCT
51  CGCCTCATGC GGCACGACCT CCGGCAAACA CCGCCAACCG AAACCCAAAC
101 AGACAGTCCG GCAAATCCAA GCCGTCCGCA TCAGCCACAT CGACCGCACA
151 CAAGGCTCGC AGGAACATCAT GCTCCACAGC CTCGGACTCA TCGGCACGCC
201 CTACAAATGG GCGCGCAGCA GCACCGCAAC CGGCTTCGAT TGCAGCGGCA
251 TGATTCAATT CGTTTACAAr AACGCCCTCA ACGTCAAGCT GCCGCGCACC
301 GCCCgcgaca TGGCGGCGGC AAGCCGsAAA ATCCCGAcA GCCGcYtCAA
351 GGCCGGCGAC CTCGTATTCT TCAACACCGG CGGCGCACAC CGCTACTCAC
401 ACGTCGGACT CTACATCGGC AACGGCGAAT TCATCCATGC CCCCAGCAGC
451 GGCAAAACCA TCAAAACCGA AAAACTCTCC ACACCGTTTT ACGCCAAAAA
501 CTACCTCGGC GCACATACTT TTTTACAGA ATGA

```

This corresponds to the amino acid sequence <SEQ ID 63; ORF 206>:

```

m206.pep.
1  MFPPDKTLFL CLSALLLASC GTTSGKHRQP KPKQTVRQIQ AVRISHIDRT
51  QGSQELMLHS LGLIGTPYKW GGSSTATGFD CSGMIQFVYK NALNVKLPRT
101 ARDMAAASRK IPDSRXKAGD LVFFNTGGAH RYSHVGLYIG NGEFIHAPSS
151 GKTIKTEKLS TPFYAKNYLG AHTFFTE*

```

The following partial DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 64>:

```

g206.seq
1  atgtttttccc ccgacaaaac ccttttctctc tgtctcggcg cactgctcct
51  cgcctcatgc ggcacgacct ccggcaaaca ccgccaacccg aaacccaaac
101 agacagtccg gcaaattccaa gccgtccgca tcagccacat cggccgcaca
151 caaggctcgc aggaactcat gctccacagc ctccggactca tcggcacgcc
201 ctacaaatgg ggcggcgagca gcaccgcaac cggcttcgac tgcagcggca
251 tgattcaatt ggttttacaaa aacgccctca acgtcaagct gccgcgcacc
301 gcccgcgaca tggcggcggc aagccgcaaa atcccgacac gccgcctcaa
351 ggccggcgac atcgtattct tcaacaccgg cggcgacacac cgctactcac
401 acgtcggact ctacatcggc aacggcggaat tcatccatgc ccccggcagc
451 ggcaaaacca tcaaaaccga aaaactctcc acaccgtttt acgcaaaaaa
501 ctaccttgga gcgcatacgt tttttacaga atga

```

- 103 -

This corresponds to the amino acid sequence <SEQ ID 65; ORF 206.ng>:

```
g206.pep
  1 MFSPDKTLFL CLGALLLASC GTTSGKHRQP KPKQTVRQIQ AVRISHIGRT
 51 QGSQELMLHS LGLIGTPYKW GGSSTATGFD CSGMIQLVYK NALNVKLPR
101 ARDMAAASRK IPDSRLKAGD IVFFNTGGAH RYSHVGLYIG NGEFIHAPGS
151 GKTIKTEKLS TPFYAKNYLG AHTFFTE*
```

ORF 206 shows 96.0% identity over a 177 aa overlap with a predicted ORF (ORF 206.ng) from *N. gonorrhoeae*:

```
m206/g206

      10      20      30      40      50      60
m206.pep MFPPDKTLFLCLSALLLASC GTTSGKHRQPKPKQTVRQIQAVRISHIDRTQGSQELMLHS
      |||||:|||||
g206      MFSPDKTLFLCLGALLLASC GTTSGKHRQPKPKQTVRQIQAVRISHIGRTQGSQELMLHS
      10      20      30      40      50      60

      70      80      90     100     110     120
m206.pep LGLIGTPYKWGGSSTATGFD CSGMIQFVYKNALNVKLPR TARDMAAASRKIPDSRXKAGD
      |||||:|||||
g206      LGLIGTPYKWGGSSTATGFD CSGMIQLVYKNALNVKLPR TARDMAAASRKIPDSRLKAGD
      70      80      90     100     110     120

      130     140     150     160     170
m206.pep LVFFNTGGAHRYSHVGLYIGNGEFIHAPSSGKTIKTEKLSTPFYAKNYLGAHTFFTEX
      :|||||:|||||
g206      IVFFNTGGAHRYSHVGLYIGNGEFIHAPSGKTIKTEKLSTPFYAKNYLGAHTFFTE
      130     140     150     160     170
```

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 66>:

```
a206.seq
  1 ATGTTTCCCC CCGACAAAAC CCTTTTCCTC TGTCTCAGCG CACTGCTCCT
 51 CGCCTCATGC GGCACGACCT CCGGCAAACA CCGCCAACCG AAACCCAAAC
101 AGACAGTCCG GCAAATCCAA GCCGTCCGCA TCAGCCACAT CGACCGCACA
151 CAAGGCTCGC AGGAACATCAT GCTCCACAGC CTCGGACTCA TCGGCACGCC
201 CTACAAATGG GGCGGCAGCA GCACCGCAAC CGGCTTCGAT TGCAGCGGCA
251 TGATTCAATT CGTTTACAAA AACGCCCTCA ACGTCAAGCT GCCGCGCACC
301 GCCCGCGACA TGGCGGCGGC AAGCCGCAAA ATCCCGGACA GCCGCCTTAA
351 GGCCGCGGAC CTCGTATTCT TCAACACCGG CGGCGCACAC CGCTACTCAC
401 ACGTCGGACT CTATATCGGC AACGGCGAAT TCATCCATGC CCCCAGCAGC
451 GGCAAAACCA TCAAAACCGA AAAACTCTCC ACACCGTTT ACGCCAAAAA
501 CTACCTCGGC GCACATACTT TCTTTACAGA ATGA
```

This corresponds to the amino acid sequence <SEQ ID 67; ORF 206.a>:

```
a206.pep
  1 MFPPDKTLFL CLSALLLASC GTTSGKHRQP KPKQTVRQIQ AVRISHIDRT
 51 QGSQELMLHS LGLIGTPYKW GGSSTATGFD CSGMIQFVYK NALNVKLPR
101 ARDMAAASRK IPDSRLKAGD LVFFNTGGAH RYSHVGLYIG NGEFIHAPSS
151 GKTIKTEKLS TPFYAKNYLG AHTFFTE*
```

m206/a206 ORFs 206 and 206.a showed a 99.4% identity in 177 aa overlap

```
      10      20      30      40      50      60
m206.pep MFPPDKTLFLCLSALLLASC GTTSGKHRQPKPKQTVRQIQAVRISHIDRTQGSQELMLHS
      |||||
a206      MFPPDKTLFLCLSALLLASC GTTSGKHRQPKPKQTVRQIQAVRISHIDRTQGSQELMLHS
      10      20      30      40      50      60
```

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	70	80	90	100	110	120
m206.pep	LGLIGTPYKWGGSSTATGFDCSGMIQFVYKNALNVKLPR	TARDMAAASRKIPDSRXKAGD				
a206	LGLIGTPYKWGGSSTATGFDCSGMIQFVYKNALNVKLPR	TARDMAAASRKIPDSRLKAGD				
	70	80	90	100	110	120
	130	140	150	160	170	
m206.pep	LVFFNTGGAHRYSHVGLYIGNGEFIHAPSSGKTIKTEKLSTPFYAKNYLGAHTFFTEX					
a206	LVFFNTGGAHRYSHVGLYIGNGEFIHAPSSGKTIKTEKLSTPFYAKNYLGAHTFFTEX					
	130	140	150	160	170	

287

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 68>:

```

m287.seq
1   ATGTTTAAAC GCAGCGTAAT CGCAATGGCT TGTATTTTGT CCCTTTCAGC
51  CTGCGGGGGC GCGGGTGGCG GATCGCCCGA TGTCAAGTCG GCGGACACGC
101 TGTCAAAACC TGCCGCCCCCT GTTGTCTCTG AAAAAAGAGC AGAGGCCAAAG
151 GAAGATGCGC CACAGGCAGG TTCTCAAGGA CAGGGCGCGC CATCCGCACA
201 AGGCAGTCAA GATATGGCGG CGGTTCGGA  AGAAAATACA GGCAATGGCG
251 GTGCGGTAAC AGCGGATAAT CCCAAAAATG AAGACGAGGT GGCACAAAAT
301 GATATGCCGC AAAATGCCGC CGGTACAGAT AGTTCGACAC CGAATCACAC
351 CCCGGATCCG AATATGCTTG CCGGAAATAT GGAAAATCAA GCAACGGATG
401 CCGGGGAATC GTCTCAGCCG GCAAACCAAC CGGATATGGC AAATGCGGCG
451 GACGGAATGC AGGGGGACGA TCCGTCGGCA GGCGGGCAAA ATGCCGGCAA
501 TACGGCTGCC CAAGGTGCAA ATCAAGCCGG AAACAATCAA GCCGCCGGTT
551 CTTCAGATCC CATCCCCGCG TCAAACCCTG CACCTGCGAA TGGCGGTAGC
601 AATTTTGGAA GGGTTGATTT GGCTAATGGC GTTTTGATTG ACGGGCCGTC
651 GCAAAATATA ACGTTGACCC ACTGTAAAGG CGATTCTTGT AGTGGCAATA
701 ATTTCTTGGA TGAAGAAGTA CAGCTAAAAT CAGAATTGGA AAAATTAAAT
751 GATGCAGACA AAATAAGTAA TTACAAGAAA GATGGGAAGA ATGATAAATT
801 TGTCGGTTTG GTTGCCGATA GTGTGCAGAT GAAGGGAATC AATCAATATA
851 TTATCTTTTA TAAACCTAAA CCCACTTCAT TTGCGCGATT TAGGCGTTCT
901 GCACGGTCGA GCGGGTCGCT TCCGGCCGAG ATGCCGCTGA TTCCCGTCAA
951 TCAGGCGGAT ACGCTGATTG TCGATGGGGA AGCGGTCAGC CTGACGGGGC
1001 ATTCGGGCAA TATCTTCGCG CCCGAAGGGA ATTACCGGTA TCTGACTTAC
1051 GGGGCGGAAA AATTGCCCGG CGGATCGTAT GCCCTTCGTG TTCAAGGCGA
1101 ACCGGCAAAA GGCGAAATGC TTGCGGGCGC GGCCGTGTAC AACGGCGAAG
1151 TACTGCATTT CCATACGGAA AACGGCCGTC CGTACCCGAC CAGGGGCAGG
1201 TTTGCCGCAA AAGTCGATTT CGGCAGCAAA TCTGTGGACG GCATTATCGA
1251 CAGCGGCGAT GATTTGCATA TGGGTACGCA AAAATTCAAA GCCGCCATCG
1301 ATGGAAACGG CTTTAAGGGG ACTTGACGG AAAATGGCAG CGGGGATGTT
1351 TCCGGAAAGT TTTACGGCCC GGCCGGCGAG GAAGTGGCGG GAAAATACAG
1401 CTATCGCCCC ACAGATGCGG AAAAGGGCGG ATTCGGCGTG TTTGCCGGCA
1451 AAAAAGAGCA GGATTGA

```

This corresponds to the amino acid sequence <SEQ ID 69; ORF 287>:

```

m287.pep
1   MFKRSVIAMA CIFALSACGG GGGGSPDVKS ADTLSKPAAP VVSEKETEAQ
51  EDAPQAGSQG QGAPSAQGSQ DMAAVSEENT GNGGAVTADN PKNEDEVAQN
101 DMPQNAAGTD SSTPNHTPDP NMLAGNMENQ ATDAGESSQP ANQPDMANAA
151 DGMQGDPPSA GGQNAGNTAA QGANQAGNNQ AAGSSDPIPA SNPAPANGGS
201 NFGRVDLANG VLIDGPSQNI TLTHCKGDSC SGNNFLDEEV QLKSEFEKLS
251 DADKISNYKK DGKNDKFVGL VADSVQMKGI NQYIIFYKPK PTSFARFRRS
301 ARSRRSLPAE MPLIPVNQAD TLIVDGEAVS LTGHSNIFA PEGNYRYLTY

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351 GAEKLPGGSY ALRVQGEPAK GEMLAGAAVY NGEVLHFHTE NGRPYPTRGR
 401 FAAKVDFGSK SVDGIIDSGD DLHMGTOQFKF AAIDGNGFKG TWTENGSGDV
 451 SGKFGYPAGE EVAGKYSYRP TDAEKGGFGV FAGKKEQD*

The following partial DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 70>:

g287.seq
 1 atgttttaaac gcagtgtgat tgcaatggct tgtatttttc cccttttcagc
 51 ctgtgggggc ggcggtggcg gatcgcccga tgtcaagtcg gcggacacgc
 101 cgtcaaaacc ggccgcccc gttgttgctg aaaatgccgg ggaaggggtg
 151 ctgccgaaag aaaagaaaga tgaggaggca gcgggcggtg cgccgcaagc
 201 cgatacgca gacgcaaccg ccggagaagg cagccaagat atggcgcaag
 251 tttcggcaga aaatacaggc aatggcggtg cggcaacaac ggacaacccc
 301 aaaaatgaag acgcgggggc gcaaaatgat atgcgcgaaa atgccgcgga
 351 atccgcaaat caaacaggga acaaccaacc cgccggttct tcagattccg
 401 cccccgcgtc aaaccctgcc cctgcgaatg gcggtagcga ttttggaagg
 451 acgaacgtgg gcaattctgt tgtgattgac ggaccgtcgc aaaatataac
 501 gttgaccacac tgtaaaggcg attcttgtaa tgggtgataat ttattggatg
 551 aagaagcacc gtcaaaatca gaatttgaaa aattaagtga tgaagaaaaa
 601 attaacgatg ataaaaaaga cgagcaacgg gagaattttg tcggtttggt
 651 tgctgacagg gtaaaaaagg atggaactaa caaatataat atcttctata
 701 cggacaaaacc acctactcgt tctgcacggt cgaggaggtc gcttccggcc
 751 gagattccgc tgattcccgat caatcaggcc gatacgtgta ttgtggatgg
 801 ggaagcggtc agcctgacgg ggcattccgg caatatcttc gcgcccggaag
 851 ggaattaccg gtatctgact tacggggcgg aaaaattgcc cggcggatcg
 901 tatgccctcc gtgtgcaagg cgaaccggca aaaggcgaaa tgcttgttgg
 951 cacggccgtg tacaacggcg aagtgtgca tttccatatg gaaaacggcc
 1001 gtccgtaccc gtccggaggc aggtttgccg caaaagtcga tttcggcagc
 1051 aaatctgtgg acggcattat cgacagcggc gatgatttgc atatgggtac
 1101 gcaaaaattc aaagccgccca tcgatggaaa cggctttaaag gggacttgga
 1151 cggaaaatgg cggcggggat gtttcgggaa ggttttacgg cccggccggc
 1201 gaggaagtgg cgggaaaata cagctatcgc ccgacagatg ctgaaaaggg
 1251 cggattcggc gtgtttgccg gcaaaaaaga tcgggattga

This corresponds to the amino acid sequence <SEQ ID 71; ORF 287.ng>:

g287.pep
 1 MFKRSVIAMA CIFPLSACGG GGGGSPDVKS ADTPSKPAAP VVAENAGEGV
 51 LPKEKKDEEA AGGAPQADTQ DATAGEGSQD MAAVSAENTG NGGAATTNDP
 101 KNEDAGAOND MPQNAAESAN QTGNNQFAGS SDSAPASNPA PANGGSDFR
 151 TNVGNVSVID GPSQNTLTH CKGDSCNGDN LLDEEAPSKS EFELKSDEEK
 201 IKRYKKDEQR ENFVGLVADR VKKDGTNKYI IFYTDKPPTR SARRSRSLPA
 251 EIPLIPVQA DTLIVDGEAV SLTGHSGNIF APEGNYRYLT YGAEKLPFGS
 301 YALRVQGEPA KGEMLVGTAV YNGEVLHFHM ENGRPYPSGG RFAAKVDFGS
 351 KSVDGIIDSG DDLHMGTOQF KAAIDGNGFK GTWTENGSGD VSGRFYGPAG
 401 EEVAGKYSYR PTDAEKGGFG VFAGKKDRD*

m287/g287 ORFs 287 and 287.ng showed a 70.1% identity in 499 aa overlap

	10	20	30	40	49
m287.pep	MFKRSVIAMACIFALSACGGGGGGSPDVKSADTSLKPAAPVVSE-----KETE				
g287	MFKRSVIAMACIFPLSACGGGGGGSPDVKSADTPSKPAAPVVAENAGEGVLPKEKKDEEA				
	10	20	30	40	50
	50	60	70	80	90
m287.pep	KEDAPQAGSQGQGAQPSAQSGDMAAVSEENTGNGGAVTADNPKNEDEVAQNDMPQNAAGT				
g287	AGGAPQADTQD--ATAGEGSQDMAAVSAENTGNGGAATTNDNPKNEDAGAQNMPQNA--				
	70	80	90	100	110

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	110	120	130	140	150	160	169
m287.pep	DSSTPNHTPDPNMLAGNMENQATDAGESSQPANQPDMANAADGMQGGDDPSAGGQNAGNTA						
g287	-----						
	170	180	190	200	210	220	229
m287.pep	AQGANQAGNNQAAGSSDPIPASNPAPANGGSNFGRVDLANGVLIDGPSQNITLTHCKGDS						
g287	-ESANQTGNNQFAGSSDSAPASNPAPANGGSDFGRTNVGNSSVVIDGPSQNITLTHCKGDS						
	120	130	140	150	160	170	
	230	240	250	260	270	280	289
m287.pep	CSGNNFLDEEVQLKSEFEKLSADADKISNYKKDGKNDKFVGLVADSVQMKGINQYIIFYKP						
g287	CNGDNLLDEEAPSKSEFEKLSDEEKIKRYKKDEQRENFVGLVADRVKKDGTNKYIIFYTD						
	180	190	200	210	220	230	
	290	300	310	320	330	340	349
m287.pep	KPTSFAFRFRSARSRRSLPAEMPLIPVNQADTLIVDGEAVSLTGHSGNIFAPEGNYRYLT						
g287	KPPT-----RSARSRRSLPAEIPVNPADTLIVDGEAVSLTGHSGNIFAPEGNYRYLT						
	240	250	260	270	280	290	
	350	360	370	380	390	400	409
m287.pep	YGAEKLPGGSYALRVQGEPAKGEMLAGAAVYNGEVLHFHTENGRPYPTRGRFAAKVDFGS						
g287	YGAEKLPGGSYALRVQGEPAKGEMLVGTAVYNGEVLHFHMENGRPYPSGGRFAAKVDFGS						
	300	310	320	330	340	350	
	410	420	430	440	450	460	469
m287.pep	KSV DGIIDSGDDLHMG TQKFKA AIDGNGFKGTWTENGSGDVSGKFYGPAGEEVAGKYSYR						
g287	KSV DGIIDSGDDLHMG TQKFKA AIDGNGFKGTWTENG GGDVSGRFGYPAGEEVAGKYSYR						
	360	370	380	390	400	410	
	470	480	489				
m287.pep	PTDAEKGFGVFAGKKEQDX						
g287	PTDAEKGFGVFAGKKDRDX						
	420	430					

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 72>:

```

a287.seq
1  ATGTTTAAAC  GCAGTGTGAT  TGCAATGGCT  TGTATTGTTG  CCCTTTTCAGC
51  CTGTGGGGGC  GCGGTGGCG  GATCGCCCGA  TGTTAAGTCG  GCGGACACGC
101  TGTCAAAC  TGCCGCCCT  GTTGTACTG  AAGATGTCGG  GGAAGAGGTG
151  CTGCCGAAAG  AAAAGAAAGA  TGAGGAGGCG  GTGAGTGGTG  CGCCGCAAGC
201  CGATACGCAG  GACGCAACCG  CCGGAAAAGG  CGGTCAAGAT  ATGGCGGCAG
251  TTTCCGCAGA  AAATACAGGC  AATGGCGGTG  CGGCAACAAC  GGATAATCCC
301  GAAAATAAAG  ACGAGGGACC  GCAAAATGAT  ATGCCGCAAA  ATGCCGCCGA
351  TACAGATAGT  TCGACACCGA  ATCACACCCC  TGCACCGAAT  ATGCCAACCA
401  GAGATATGGG  AAACCAAGCA  CCGATGCGG  GGAATCGGC  ACAACCGCA
451  AACCAACCGG  ATATGGCAAA  TCGCGCGGAC  GGAATGCAGG  GGGACGATCC
501  GTCGGCAGGG  GAAAATGCCG  GCAATACGGC  AGATCAAGCT  GCAAATCAAG
551  CTGAAAACAA  TCAAGTCGGC  GGCTCTCAAA  ATCCTGCCTC  TTCAACCAAT
601  CCTAACGCCA  CGAATGGCGG  CAGCGATTTT  GGAAGGATAA  ATGTAGCTAA
651  TGGCATCAAG  CTTGACAGCG  GTTCGGAAAA  TGTAACGTTG  ACACATTGTA
701  AAGACAAAGT  ATGCGATAGA  GATTTCTTAG  ATGAAGAAGC  ACCACCAAAA
751  TCAGAATTTG  AAAAATTAAG  TGATGAAGAA  AAAATTAATA  AATATAAAAA
801  AGACGAGCAA  CGAGAGAATT  TTGTCGGTTT  GGTGCTGAC  AGGGTAGAAA

```

a287.pwp

m287/a287 ORFs 287 and 287.a showed a 77.2% identity in 501 aa overlap

m287.pep KP--TSFARFRRSARRSLPAEMPLIPVNQADTLIVDGEAVSLTGHSNIFAPEGNYRY
 | : | | | | | | | | | | | | | | | | | | | | | |

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a287      KSASSSSARFRRSARSRRSLPAEMPLIPVNQADTLIVDGEAVSLTGHSGNIFAPEGNYRY
           300      310      320      330      340      350

m287.pep  350      360      370      380      390      400
           LTYGAEKLPGGSYALRVQGEPAKGEMLAGAAVYNGEVLHFHTENGRPYPTRGRFAAKVDF
           ||||| ||||| ||||| |||||:||||| ||||| ||||| |: |||||
a287      LTYGAEKLSGGSYALSVQGEPAKGEMLAGTAVYNGEVLHFHFMENGRPSPSGGRFAAKVDF
           360      370      380      390      400      410

m287.pep  410      420      430      440      450      460
           GSKSVDGIIDSGDDLHMGTKQFKAAIDGNGFKGTWTENGSGDVS GKFGYPAGEEVAGKYS
           ||||| ||||| ||||| |||||:||||| ||||| |||||:||||| |||||
a287      GSKSVDGIIDSGDDLHMGTKQFKAVIDGNGFKGTWTENGSGDVS GRFYGPAGEEVAGKYS
           420      430      440      450      460      470

m287.pep  470      480      489
           YRPTDAEKGGFGVFAGKKEQDX
           ||||| ||||| |||||
a287      YRPTDAEKGGFGVFAGKKEQDX
           480      490

```

406

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 74>:

m406.seq

```

1  ATGCAAGCAC GGCTGCTGAT ACCTATTCTT TTTTCAGTTT TTATTTTATC
51  CGCCTGCGGG AACTGACAG GTATTCCATC GCATGCGGGA GGTAACGCT
101 TTGCGGTCGA ACAAGAACTT GTGGCCGCTT CTGCCAGAGC TGCCGTAA
151 GACATGGATT TACAGGCATT ACACGGACGA AAAGTTGCAT TGTACATTGC
201 CACTATGGGC GACCAAGGTT CAGGCAGTTT GACAGGGGGT CGCTACTCCA
251 TTGATGCACT GATTCGTGGC GAATACATAA ACAGCCCTGC CGTCCGTACC
301 GATTACACCT ATCCACGTTA CGAAACCACC GCTGAAACAA CATCAGGCGG
351 TTTGACAGGT TTAACCACTT CTTTATCTAC ACTTAATGCC CCTGCACTCT
401 CTCGCACCCA ATCAGACGGT AGCGGAAGTA AAAGCAGTCT GGGCTTAAAT
451 ATTGGCGGGA TGGGGGATTA TCGAAATGAA ACCTTGACGA CTAACCCGCG
501 CGACACTGCC TTCTTTCCC ACTTGGTACA GACCGTATTT TTCCTGCGCG
551 GCATAGACGT TGTTTCTCCT GCCAATGCCG ATACAGATGT GTTTATTAAC
601 ATCGACGTAT TCGGAACGAT ACGCAACAGA ACCGAAATGC ACCTATACAA
651 TGCCGAAACA CTGAAAGCCC AAACAAACT GGAATATTC GCAGTAGACA
701 GAACCAATAA AAAATTGCTC ATCAAACCAA AAACCAATGC GTTTGAAGCT
751 GCCTATAAAG AAAATTACGC ATTGTGGATG GGGCCGTATA AAGTAAGCAA
801 AGGAATTAAA CCGACGGAAG GATTAATGGT CGATTCTCC GATATCCGAC
851 CATACGGCAA TCATACGGGT AACTCCGCCC CATCCGTAGA GGCTGATAAC
901 AGTCATGAGG GGTATGGATA CAGCGATGAA GTAGTGCGAC AACATAGACA
951 AGGACAACCT TGA

```

This corresponds to the amino acid sequence <SEQ ID 75; ORF 406>:

m406.pep

```

1  MQARLLIPIL FSVFILSACG TLTGIPSHGG GKRFAVEQEL VAASARAAVK
51  DMDLQALHGR KVALYIATMG DQSGSLTGG RYSIDALIRG EYINSPAVRT
101 DYTPRYETT AETTSGLTGT LTSLSTLNA PALSRTQSDG SGSKSSLGLN
151 IGGMGDYRNE TLTTNPRDTA FLSHLVQTVF FLRGIDVVSP ANADTDVFIN
201 IDVFGTIRNR TEMHLYNAET LKAQTKLEYF AVDRTNKKLL IKPKTNAFEA
251 AYKENYALWM GPYKVSKEIK PTEGLMVDFS DIRPYGNHTG NSAPSVEADN
301 SHEGYGYSDE VVRQHRQGP *

```

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The following partial DNA sequence was identified in *N. gonorrhoeae* <SEQ ID 76>:

g406.seq

```

1   ATGCGGGCAC GGCTGCTGAT ACCTATTCTT TTTTCAGTTT TTATTTTATC
51  CGCCTGCGGG ACACTGACAG GTATTCCATC GCATGGCGGA GGCAAACGCT
101 TCGCGGTCGA ACAAGAACTT GTGGCCGCTT CTGCCAGAGC TGCCGTAAAC
151 GACATGGATT TACAGGCATT ACACGGACGA AAAGTTGCAT TGTACATTGC
201 AACTATGGGC GACCAAGGTT CAGGCAGTTT GACAGGGGGT CGCTACTCCA
251 TTGATGCACT GATTGCGGCG GAATACATAA ACAGCCCTGC CGTCCGCACC
301 GATTACACCT ATCCGCGTTA CGAAACCACC GCTGAAACAA CATCAGGCGG
351 TTTGACGGGT TTAACCACTT CTTTATCTAC ACTTAATGCC CCTGCACTCT
401 CGCGCACCCA ATCAGACGGT AGCGGAAGTA GGAGCAGTCT GGGCTTAAAT
451 ATTGGCGGGA TGGGGGATTA TCGAAATGAA ACCTTGACGA CCAACCCGCG
501 CGACACTGCC TTTCTTTCCC ACTTGGTGCA GACCGTATTT TTCCTGCGCG
551 GCATAGACGT TGTTTCTCCT GCCAATGCCG ATACAGATGT GTTTATTAAC
601 ATCGACGTAT TCGGAACGAT ACGCAACAGA ACCGAAATGC ACCTATACAA
651 TGCCGAAACA CTGAAAGCCC AAACAAACT GGAATATTC GCAGTAGACA
701 GAACCAATAA AAAATTGCTC ATCAAACCCA AAACCAATGC GTTTGAAGCT
751 GCCTATAAAG AAAATTACGC ATTGTGGATG GGGCCGTATA AAGTAAGCAA
801 AGGAATCAAA CCGACGGAAG GATTGATGGT CGATTTCTCC GATATCCAAC
851 CATACGGCAA TCATACGGGT AACTCCGCC CATCCGTAGA GGCTGATAAC
901 AGTCATGAGG GGTATGGATA CAGCGATGAA GCAGTGCAC AACATAGACA
951 AGGGCAACCT TGA

```

This corresponds to the amino acid sequence <SEQ ID 77; ORF 406.ng>:

g406.pep

```

1   MRARLLIPIL FSVFILSACG TLTGIPSHGG GKRFAVEQEL VAASARAAVK
51  DMDLQALHGR KVALYIATMG DQSGSGLTGG RYSIDALIRG EYINSPAVRT
101 DYTPRYETT AETTSGLTG LTSLSTLNA PALSRQSDG SGSRSSLGLN
151 IGGMGDYRNE TLTTNPRDTA FLSHLVQTVF FLRGIDVVSP ANADTDVFIN
201 IDVFGTIRNR TEMHLYNAET LKAQTKLEYF AVDRTNKKLL IKPKTNAFEA
251 AYKENYALWM GPYKVSKEIK PTEGLMVDFS DIQPYGNHTG NSAPSV EADN
301 SHEGYGYSDE AVRQHRQGP *

```

ORF 406.ng shows 98.8% identity over a 320 aa overlap with a predicted ORF (ORF406.a) from *N. gonorrhoeae*:

g406/m406

	10	20	30	40	50	60
g406.pep	MRARLLIPILFSVFILSACGTLTGIPSHGGGKRFVEQELVAASARA AVKMDLQALHGR					
m406	MQARLLIPILFSVFILSACGTLTGIPSHGGGKRFVEQELVAASARA AVKMDLQALHGR					
	10	20	30	40	50	60
	70	80	90	100	110	120
g406.pep	KVALYIATMGDQSGSGLTGGRYSIDALIRGEYINSPAVRTDYTPRYETTAETTSGLTG					
m406	KVALYIATMGDQSGSGLTGGRYSIDALIRGEYINSPAVRTDYTPRYETTAETTSGLTG					
	70	80	90	100	110	120
	130	140	150	160	170	180
g406.pep	LTSLSTLNAPALSRQSDGSGSRSSLGLNIGGMGDYRNETLTNPRDTAFLSHLVQTVF					
m406	LTSLSTLNAPALSRQSDGSGSKSSLGLNIGGMGDYRNETLTNPRDTAFLSHLVQTVF					
	130	140	150	160	170	180
	190	200	210	220	230	240

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```

g406.pep  FLRGIDVVSPANADTDVFINIDVFGTIRNRTEMHLYNAETLKAQTKLEYFAVDRTNKKLL
          |||
m406      FLRGIDVVSPANADTDVFINIDVFGTIRNRTEMHLYNAETLKAQTKLEYFAVDRTNKKLL
          190      200      210      220      230      240

          250      260      270      280      290      300
g406.pep  IKPKTNAFEAAAYKENYALWMGPYKVSKGIKPTEGLMVDFSIDIQPYGNHTGNSAPSVEADN
          |||
m406      IKPKTNAFEAAAYKENYALWMGPYKVSKGIKPTEGLMVDFSIDIQPYGNHTGNSAPSVEADN
          250      260      270      280      290      300

          310      320
g406.pep  SHEGYGYSDEAVRQHRQGQPX
          |||
m406      SHEGYGYSDEVVRQHRQGQPX
          310      320

```

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 78>:

```

a406.seq
1  ATGCAAGCAC  GGCTGCTGAT  ACCTATTCTT  TTTTCAGTTT  TTATTTTATC
51  CGCCTGCGGG  ACACTGACAG  GTATTCCATC  GCATGGCGGA  GGTAAACGCT
101  TCGCGGTCGA  ACAAGAACTT  GTGGCCGCTT  CTGCCAGAGC  TGCCGTTAAA
151  GACATGGATT  TACAGGCATT  ACACGGACGA  AAAGTTGCAT  TGTACATTGC
201  AACTATGGGC  GACCAAGGTT  CAGGCAGTTT  GACAGGGGGT  CGCTACTCCA
251  TTGATGCACT  GATTCGTGGC  GAATACATAA  ACAGCCCTGC  CGTCCGTACC
301  GATTACACCT  ATCCACGTTA  CGAAACCACC  GCTGAAACAA  CATCAGGCGG
351  TTTGACAGGT  TTAACCACTT  CTTTATCTAC  ACTTAATGCC  CCTGCACTCT
401  CGCGCACCCA  ATCAGACGGT  AGCGGAAGTA  AAAGCAGTCT  GGGCTTAAAT
451  ATTGCGCGGA  TGGGGGATTA  TCGAAATGAA  ACCTTGACGA  CTAACCCGCG
501  CGACACTGCC  TTTCTTTCCC  ACTTGGTACA  GACCGTATTT  TTCCTGCGCG
551  GCATAGACGT  TGTTTCTCCT  GCCAATGCCG  ATACGGATGT  GTTTATTAAC
601  ATCGACGTAT  TCGGAACGAT  ACGCAACAGA  ACCGAAATGC  ACCTATACAA
651  TGCCGAAACA  CTGAAAGCCC  AAACAAAAC  GGAATATTTT  GCAGTAGACA
701  GAACCAATAA  AAAATTGCTC  ATCAAACCAA  AAACCAATGC  GTTTGAAGCT
751  GCCTATAAAG  AAAATTACGC  ATTGTGGATG  GGACCGTATA  AAGTAAGCAA
801  AGGAATTAAA  CCGACAGAAG  GATTAATGGT  CGATTCTCTC  GATATCCAAC
851  CATACGGCAA  TCATATGGGT  AACTCTGCCC  CATCCGTAGA  GGCTGATAAC
901  AGTCATGAGG  GGTATGGATA  CAGCGATGAA  GCAGTGCGAC  GACATAGACA
951  AGGGCAACCT  TGA

```

This corresponds to the amino acid sequence <SEQ ID 79; ORF 406.a>:

```

a406.pep
1  MQARLLIPIL  FSVFILSACG  TLTGIPSHGG  GKRFAVEQEL  VAASARAAVK
51  DMDLQALHGR  KVALYIATMG  DQSGSGLTGG  RYSIDALIRG  EYINSPAVRT
101  DYTYPRYETT  AETTSGLTGG  LTSLSTLNA  PALSRTQSDG  SGSSSLGLN
151  IGGMGDYRNE  TLTTNPRDTA  FLSHLVQTVF  FLRGIDVVSP  ANADTDVFIN
201  IDVFGTIRNR  TEMHLYNAET  LKAQTKLEYF  AVDRTNKKLL  IKPKTNAFEA
251  AYKENYALWM  GPYKVSKGIK  PTEGLMVDFS  DIQPYGNHMG  NSAPSVEADN
301  SHEGYGYSDE  AVRRHRQGQP  *

```

m406/a406 ORFs 406 and 406.a showed a 98.8% identity in 320 aa overlap

```

          10      20      30      40      50      60
m406.pep  MQARLLIPILFSVFILSACGTLTGIPSHGGGKRFAVEQELVAASARAAVKDMDLQALHGR
          |||
a406      MQARLLIPILFSVFILSACGTLTGIPSHGGGKRFAVEQELVAASARAAVKDMDLQALHGR
          10      20      30      40      50      60

          70      80      90      100     110     120
m406.pep  KVALYIATMGDQSGSGLTGGRYSIDALIRGEYINSPAVRTDYTYPRYETTAETTSGLTGG
          |||

```

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a406	KVALYIATMGDQGSGLTGGRYSIDALIRGEYINSPAVRTDYTPRYETTAETTSGGLTG
	70 80 90 100 110 120
m406.pep	130 140 150 160 170 180
	LTTSLSTLNAPALSRTQSDGSGSKSSLGLNIGMGDYRNETLTTNPRDTAFLSHLVQTVF
a406	
	LTTSLSTLNAPALSRTQSDGSGSKSSLGLNIGMGDYRNETLTTNPRDTAFLSHLVQTVF
	130 140 150 160 170 180
m406.pep	190 200 210 220 230 240
	FLRGIDVVS PANADTDVFINIDVFGTIRNRTEMHLYNAETLKAQTKLEYFAVDRTNKKLL
a406	
	FLRGIDVVS PANADTDVFINIDVFGTIRNRTEMHLYNAETLKAQTKLEYFAVDRTNKKLL
	190 200 210 220 230 240
m406.pep	250 260 270 280 290 300
	IKPKTNAFEAAAYKENYALWMGPYKVS KG IKPTEGLMVD FSDIRPYGNHTGNSAPSVEADN
a406	
	IKPKTNAFEAAAYKENYALWMGPYKVS KG IKPTEGLMVD FSDIQPYGNHMGNSAPSVEADN
	250 260 270 280 290 300
m406.pep	310 320
	SHEGYGYSDEVVRQHRQGQPX
a406	
	SHEGYGYSDEAVRRHRQGQPX
	310 320

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 80>:

m726.seq

1	ATGACCATCT	ATTTCAAAAA	CGGCTTTTAC	GACGACACAT	TGGGCGGCAT
51	CCCCGAAGGC	GCGGTTGCCG	TCCGCGCCGA	AGAATACGCC	GCCCTTTTGG
101	CAGGACAGGC	GCAGGGCGGG	CAGATTGCCG	CAGATTCCGA	CGGCCGCCCC
151	GTTTTAACC	CGCCGCGCCC	GTCCGATTAC	CACGAATGGG	ACGGCAAAAA
201	ATGGAAAATC	AGCAAAGCCG	CCGCCGCCG	CCGTTTCGCC	AAACAAAAAA
251	CCGCCTTGGC	ATTCCGCCTC	GCGGAAAAGG	CGGACGAACT	CAAAAACAGC
301	CTCTTGCGG	GCTATCCCCA	AGTGGAATC	GACAGCTTTT	ACAGGCAGGA
351	AAAAGAAGCC	CTCGCGCGGC	AGGCGGACAA	CAACGCCCCG	ACCCCGATGC
401	TGGCGCAAAT	CGCCGCGCGA	AGGGGCGTGG	AATTGGACGT	TTTGATTGAA
451	AAAGTTATCG	AAAAATCCGC	CCGCCTGGCT	GTTGCCGCCG	GCGCGATTAT
501	CGGAAAGCGT	CAGCAGCTCG	AAGACAAATT	GAACACCATC	GAAACCGCGC
551	CCGGATTGGA	CGCGCTGGAA	AAGGAAATCG	AAGAATGGAC	GCTAAACATC
601	GGCTGA				

This corresponds to the amino acid sequence <SEQ ID 81; ORF 726>:

m726.pep

1	MTIYFKNGFY	DDTLGGIPEG	AVAVRAEEYA	ALLAGQAQGG	QIAADSDGRP
51	VLTPPPPSDY	HEWDGKKWKI	SKAAAAARFA	KQKTALAFRL	AEKADELKNS
101	LLAGYPQVEI	DSFYRQEKEA	LARQADNNAP	TPMLAQIAAA	RGVELDLVIE
151	KVIEKSARLA	VAAGAIIGKR	QQLLEDKLNIT	ETAPGLDALE	KEIEEWTINI
201	G*				

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 82>:

m907-2.seq

1	ATGAGAAAAAC	CGACCGATAC	CCTACCCGTT	AATCTGCAAC	GCCGCCGCCT
51	GTTGTGTGCC	GCCGGTGCGT	TGTTGCTCAG	TCCTCTGGCG	CACGCCGGCG
101	CGCAACGTGA	GGAAACGCTT	GCCGACGATG	TGGCTTCCGT	GATGAGGAGT

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```

151 TCTGTCGGCA GCGTCAATCC GCCGAGGCTG GTGTTTGACA ATCCGAAAGA
201 GGGCGAGCGT TGGTTGTCTG CCATGTCGGC ACGTTTGGCA AGGTTTCGTCC
251 CCGAGGAGGA GGAGCGGCGC AGGCTGCTGG TCAATATCCA GTACGAAAGC
301 AGCCGGGCGG GTTTGGATAC GCAGATTGTG TTGGGGCTGA TTGAGGTGGA
351 AAGCGCGTTC CGCCAGTATG CAATCAGCGG TGTCGGCGCG CGCGGCCTGA
401 TGCAGGTTAT GCCGTTTTTG AAAAATAACA TCGGCAAACC GGCGCACAAC
451 CTGTTTCGACA TCCGCACCAA CCTGCGTTAC GGCTGTACCA TCCTGCGCCA
501 TTACCGGAAT CTTGAAAAAG GCAACATCGT CCGCGCGCTT GCCCGCTTTA
551 ACGGCAGCTT GGGCAGCAAT AAATATCCGA ACGCCGTTTT GGGCGCGTGG
601 CGCAACCGCT GGCAGTGGCG TTGA

```

This corresponds to the amino acid sequence <SEQ ID 83; ORF 907-2>:

m907-2.pep

```

1 MRKPTDTLPV NLQRRRLCA AGALLLSPLA HAGAQREETL ADDVASVMRS
51 SVGSVNPPRL VFDNPKEGER WLSAMSARLA RFVPEEEERR RLLVNIQYES
101 SRAGLDTQIV LGLIEVESAF RQYAISGVGA RGLMQVMPFW KNYIGKPAHN
151 LFDIRTNLRY GCTILRHYRN LEKGNIVRAL ARFNGSLGSN KYPNAVLGAW
201 RNRWQWR*

```

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 84>:

m953.seq

```

1 ATGAAAAAAA TCATCTTCGC CGCACTCGCA GCCGCCGCCA TCAGTACTGC
51 CTCCGCCGCC ACCTACAAAG TGGACGAATA TCACGCCAAC GCCCGTTTCG
101 CCATCGACCA TTTCAACACC AGCACCAACG TCGGCGGTTT TTACGGTCTG
151 ACCGGTTCCG TCGAGTTCGA CCAAGCAAAA CGCGACGGTA AAATCGACAT
201 CACCATCCCC ATTGCCAACC TGCAAAGCGG TTCGCAACAC TTTACCGACC
251 ACCTGAAATC AGCCGACATC TTCGATGCCG CCCAATATCC GGACATCCGC
301 TTTGTTTCCA CCAAATTCAA CTTCAACGGC AAAAATACTG TTTCCGTTGA
351 CGGCAACCTG ACCATGCACG GCAAAACCGC CCCCCTCAAA CTCAAAGCCG
401 AAAAATTCAA CTGCTACCAA AGCCCGATGG AGAAAACCGA AGTTTGTGGC
451 GGCGACTTCA GCACCACCAT CGACCGCACC AAATGGGGCA TGGACTACCT
501 CGTTAACGTT GGTATGACCA AAAGCGTCCG CATCGACATC CAAATCGAGG
551 CAGCCAAACA ATAA

```

This corresponds to the amino acid sequence <SEQ ID 85; ORF 953>:

m953.pep

```

1 MKKIIFAALA AAAISTASAA TYKVDEYHAN ARFAIDHFNT STNVGGFYGL
51 TGSVEFDQAK RDGKIDITIP IANLQSGSQH FTDHLKSADI FDAAQYPDIR
101 FVSTKFNENG KKLVSVDGNL TMHGKTAPVK LKAEKFNCYQ SPMEKTEVCG
151 GDFSTTIDRT KWGMDYLVNV GMTKSVRIDI QIEAAKQ*

```

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 86>:

orf1-1.seq

```

1 ATGAAAACAA CCGACAAACG GACAACCGAA ACACACCGCA AAGCCCCGAA
51 AACCGGCCGC ATCCGCTTCT CGCCTGCTTA CTTAGCCATA TGCCTGTCGT
101 TCGGCATTCT TCCCCAAGCC TGGCGGGGAC ACACTTATTT CGGCATCAAC
151 TACCAATACT ATCGCGACTT TGCCGAAAAT AAAGGCAAGT TTGCAGTCGG
201 GGCGAAAGAT ATTGAGGTTT ACAACAAAAA AGGGGAGTTG GTCGGCAAAT
251 CAATGACAAA AGCCCCGATG ATTGATTTTT CTGTGGTGTC GCGTAACGGC
301 GTGCGGGCAT TGGTGGGCGA TCAATATATT GTGAGCGTGG CACATAACGG
351 CGGCTATAAC AACGTTGATT TTGGTGCGGA AGGAAGAAAT CCCGATCAAC
401 ATCGTTTTAC TTATAAAATT GTGAAACGGA ATAATTATAA AGCAGGGACT
451 AAAGGCCATC CTTATGGCGG CGATTATCAT ATGCCGCGTT TGCATAAATT
501 TGTCACAGAT GCAGAACCTG TTGAAATGAC CAGTTATATG GATGGGCGGA

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551 AATATATCGA TCAAATAAT TACCCTGACC GTGTTCTGAT TGGGGCAGGC
601 AGGCAATATT GGCGATCTGA TGAAGATGAG CCCAATAACC GCGAAAGTTC
651 ATATCATATT GCAAGTGCGT ATTCTTGGCT CGTTGGTGGC AATACCTTTG
701 CACAAAATGG ATCAGGTGGT GGCACAGTCA ACTTAGGTAG TGAAAAAATT
751 AAACATAGCC CATATGGTTT TTTACCAACA GGAGGCTCAT TTGGCGACAG
801 TGGCTCACCA ATGTTTATCT ATGATGCCCA AAAGCAAAAG TGGTTAATTA
851 ATGGGGTATT GCAAACGGGC AACCCCTATA TAGGAAAAG CAATGGCTTC
901 CAGCTGGTTC GTAAAGATTG GTTCTATGAT GAAATCTTTG CTGGAGATAC
951 CCATTCAGTA TTCTACGAAC CACGTCAAAA TGGGAAATAC TCTTTTACG
1001 ACGATAATAA TGGCACAGGA AAAATCAATG CCAAACATGA ACACAATTCT
1051 CTGCCTAATA GATTAAAAAC ACGAACCGTT CAATTGTTTA ATGTTTCTTT
1101 ATCCGAGACA GCAAGAGAAC CTGTTTATCA TGCTGCAGGT GGTGTCAACA
1151 GTTATCGACC CAGACTGAAT AATGGAGAAA ATATTTCTCT TATTGACGAA
1201 GGAAAAGGCG AATTGATACT TACCAGCAAC ATCAATCAAG GTGCTGGAGG
1251 ATTATATTTC CAAGGAGATT TTACGGTCTC GCCTGAAAAT AACGAACTT
1301 GGCAAGGCGC GGGCGTTCAT ATCAGTGAAG ACAGTACCGT TACTTGGAAG
1351 GTAAACGGCG TGGCAAACGA CCGCTGTCC AAAATCGGCA AAGGCACGCT
1401 GCACGTTCAA GCCAAAGGGG AAAACCAAGG CTCGATCAGC GTGGGCGACG
1451 GTACAGTCAT TTTGGATCAG CAGGCAGACG ATAAAGGCAA AAAACAAGCC
1501 TTTAGTGAAA TCGGCTTGGT CAGCGGCAGG GGTACGGTGC AACTGAATGC
1551 CGATAATCAG TTCAACCCCG ACAACTCTA TTTGCGCTTT CGCGGCGGAC
1601 GTTTGGATTT AAACGGGCAT TCGCTTTCGT TCCACCGTAT TCAAAATACC
1651 GATGAAGGGG CGATGATTGT CAACCACAAT CAAGACAAAG AATCCACCGT
1701 TACCATTACA GGCAATAAAG ATATTGCTAC AACC GGCAAT AACAACAGCT
1751 TGGATAGCAA AAAAGAAATT GCCTACAACG GTTGGTTTGG CGAGAAAGAT
1801 ACGACCAAAA CGAACGGGCG GCTCAACCTT GTTACCAGC CCGCCGACGA
1851 AGACCGCACC CTGCTGCTTT CCGGCGGAAC AAATTTAAAC GGCAACATCA
1901 CGCAACAAA CGGCAACTG TTTTTCAGCG GCAGACCAAC ACCGCACGCC
1951 TACAATCAT TAAACGACCA TTGGTTCGCA AAAGAGGGCA TTCCTCGCGG
2001 GGAAATCGTG TGGGACAACG ACTGGATCAA CCGCACATTT AAAGCGGAAA
2051 ACTTCCAAAT TAAAGGCGGA CAGGCGGTGG TTTCCCGCAA TGTTGCCAAA
2101 GTGAAAGGCG ATTGGCATT T GAGCAATCAC GCCCAAGCAG TTTTGGTGT
2151 CGCACCGCAT CAAAGCCACA CAATCTGTAC ACGTTCGGAC TGGACGGGTC
2201 TGACAAATTG TGTCGAAAAA ACCATTACCG ACGATAAAGT GATTGCTTCA
2251 TTGACTAAGA CCGACATCAG CGGCAATGTC GATCTTGCCG ATCACGCTCA
2301 TTTAAATCTC ACAGGGCTTG CCACACTCAA CGGCAATCTT AGTGCAAATG
2351 GCGATACACG TTATACAGTC AGCCACAACG CCACCCAAAA CGGCAACCTT
2401 AGCCTCGTGG GCAATGCCCA AGCAACATTT AATCAAGCCA CATTAAACGG
2451 CAACACATCG GCTTCGGGCA ATGCTTCATT TAATCTAAGC GACCACGCCG
2501 TACAAAACGG CAGTCTGACG CTTTCCGGCA ACGCTAAGGC AAACGTAAGC
2551 CATTCCGCAC TCAACGGTAA TGTCTCCCTA GCCGATAAGG CAGTATTCCA
2601 TTTTGAAAGC AGCCGCTTTA CCGGACAAAT CAGCGGCGGC AAGGATACGG
2651 TATTACACTT AAAAGACAGC GAATGGACGC TGCCGTCAGG CACGGAATTA
2701 GGCAATTTAA ACCTTGACAA CGCCACCATT ACACTCAATT CCGCCTATCG
2751 CCACGATGCG GCAGGGGCGC AAACCGGCAG TCGGACAGAT GCGCCGCGCC
2801 GCCGTTCCGC CCGTTCGCGC CGTTCCTTAT TATCCGTTAC ACCGCCAAT
2851 TCGGTAGAAAT CCCGTTTCAA CACGCTGACG GTAAACGGCA AATTGAACGG
2901 TCAGGGAACA TTCCGCTTTA TGTCGGAAC TTTGCGCTAC CGCAGCGACA
2951 AATTGAAGCT GGCGGAAAGT TCCGAAGGCA CTTACACCTT GGCGGTCAAC
3001 AATACCGGCA ACGAACCTGC AAGCCTCGAA CAATTGACGG TAGTGGAAGG
3051 AAAAGACAAC AAACCGCTGT CCGAAAACCT TAATTTCAAC CTGCAAAACG
3101 AACACGTCGA TGCCGGCGCG TGGCGTTACC AACTCATCCG CAAAGACGGC
3151 GAGTTCCGCC TGCATAATCC GGTCAAAGAA CAAGAGCTTT CCGACAAACT
3201 CGGCAAGGCA GAAGCCAAAA AACAGGCGGA AAAAGACAAC GCGCAAAGCC
3251 TTGACGCGCT GATTGCGGCC GGGCGCGATG CCGTCGAAAA GACAGAAAGC
3301 GTTGCCGAAC CGGCCCGGCA GGCAGGCGGG GAAATGTCTG GCATTATGCA
3351 GCGGGAGGAA GAGAAAAAAC GGGTGCAGGC GGATAAAGAC ACCGCCTTGG
3401 CGAAACAGCG CGAAGCGGAA ACCCGGCCGG CTACCACCGC CTTCCCCCGC
3451 GCGCGCCGCG CCCGCCGGGA TTTGCCGCAA CTGCAACCCC AACCGCAGCC
3501 CCAACCGCAG CGCGACCTGA TCAGCCGTTA TGCCAATAGC GGTTTGAGTG
3551 AATTTTCCGC CACGCTCAAC AGCGTTTTTC CCGTACAGGA CGAATTAGAC
3601 CGCGTATTTG CCGAAGACCG CCGCAACGCC GTTTGGACAA GCGGCATCCG
3651 GGACACCAAA CACTACCGTT CGCAAGATTT CCGCGCCTAC CGCCAACAAA

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3701 CCGACCTGCG CCAAATCGGT ATGCAGAAAA ACCTCGGCAG CGGGCGCGTC
3751 GGCATCCTGT TTTTCGCACAA CCGGACCGAA AACACCTTCG ACGACGGCAT
3801 CCGCAACTCG GCACGGCTTG CCCACGGCGC CGTTTTCGGG CAATACGGCA
3851 TCGACAGGTT CTACATCGGC ATCAGCGCGG GCGCGGGTTT TAGCAGCGGC
3901 AGCCTTTCAG ACGGCATCGG AGGCAAAATC CGCCGCCGCG TGCTGCATTA
3951 CGGCATTTCAG GCACGATACC GCGCCGGTTT CCGCGGATTC GGCATCGAAC
4001 CGCACATCGG CGCAACGCGC TATTTTCGTCC AAAAAGCGGA TTACCGCTAC
4051 GAAAACGTCA ATATCGCCAC CCCCGGCCTT GCATTCAACC GCTACCGCGC
4101 GGGCATTAAAG GCAGATTATT CATTCAAACC GGCGCAACAC ATTTCCATCA
4151 CGCCTTATTT GAGCCTGTCC TATACCGATG CCGCTTCGGG CAAAGTCCGA
4201 ACACGCGTCA ATACCGCCGT ATTGGCTCAG GATTTCGGCA AAACCCGCAG
4251 TGCGGAATGG GGCCTAAACG CCGAAATCAA AGGTTTCACG CTGTCCCTCC
4301 ACGCTGCCGC CGCCAAAGGC CCGCAACTGG AAGCGCAACA CAGCGCGGGC
4351 ATCAAATTAG GCTACCGCTG GTAA

```

This corresponds to the amino acid sequence <SEQ ID 87; ORF orf1-1>:

orf1-1.pep

```

1 MKTTDKRTE THRKAPKTGR IRFSPAYLAI CLSFGILPQA WAGHTYFGIN
51 YQYYRDAEN KGKFAVGAKD IEVYNKKGEL VGKSMTKAPM IDFSVVS RNG
101 VAALVGDQYI VSAHNGGYN NVDFGAEGRN PDQHRFTYKI VKRNNYKAGT
151 KGHYPYGGDYH MPRLHKFVTD AEPVEMTSYM DGRKYIDQNN YPDRVRIGAG
201 RQYWRSEDE PNNRESSYHI ASAYSWLVGG NTFAQNGSGG GTVNLGSEKI
251 KHSPYGFLLPT GGSFGDSGSP MFIYDAQKQK WLINGVLQTG NPYIGKSNGF
301 QLVKRDWFDY EIFAGDTHSV FYEPRQNGKY SFNDNNGTG KINAKHEHNS
351 LPNRLKTRTV QLFNVSLSET AREPVYHAA GVNYSYRPRLN NGENISFIDE
401 KGELILTSN INQAGGLYF QGDFTVSPEN NETWQAGAVH ISEDSTVTWK
451 VNGVANDRLS KIGKGTLLHVQ AKGENQGSIS VGDGTVILDQ QADDKGGKQA
501 FSEIGLVSGR GTVQLNADNQ FNPDKLYFGF RGGRLDLNGH SLSFHRIQNT
551 DEGAMIVNHN QDKESTVTIT GNKDIATTGN NNSLDSKKEI AYNGWFGKED
601 TTKTNGRLNL VYQPAEDRT LLLSGGTNLN GNITQTNGKL FFSGRPTPHA
651 YNHLNDHWSQ KEGIPRGEIV WDNDWINRTF KAENFQIKGG QAVVSRNVAK
701 VKGDWHLNSH AQAVFGVAPH QSHTICTRSD WTGLTNCVEK TITDDKVIAS
751 LTKTDISGNV DLADHAHLNL TGLATLNGNL SANGDTRYTV SHNATQNGNL
801 SLVGNAQATF NQATLNGNTS ASGNASENLS DHAVQNGSLT LSGNAKANVS
851 HSA LNGNVSL ADKAVFHFES SRFTGQISGG KDTALHLKDS EWTLPSTEL
901 GNLNLNDNATI TLNSAYRHDA AGAQTGSATD APRRRSRRSR RSLLSVTPPT
951 SVESRFNTLT VNGKLNQGT FREMSELFY RSDKLKLAES SEGTYTLAVN
1001 NTGNEPASLE QLTVEGKDN KPLSENLFNT LQNEHVDAGA WRYQLIRKDG
1051 EFRLHNPVKE QELSDKLGA EAKKQAEKDN AQSLDALIAA GRDAVEKTES
1101 VAEPARQAGG ENVGIMQAE EKRVQADKD TALAKQREAE TRPATTAFFR
1151 ARRARRDLPQ LQPQPQPQPQ RDLISRYANS GLSEFSATLN SVFAVQDELD
1201 RVFAEDRRNA VWTSGIRDTK HYRSQDFRAY RQQTDLRQIG MQKNLGSGRV
1251 GILFSHN RTE NTFDDGIGNS ARLAHGAVFG QYGIDRFYIG ISAGAGFSSG
1301 SLSDGIGGKI RRRVLHYGIQ ARYRAGFGGF GIEPHIGATR YFVQKADYRY
1351 ENVNIATPGL AFNRYRAGIK ADYSFKPAQH ISITPYLSLS YTDAAAGKVR
1401 TRVNTAVLAQ DFGKTRSAEW GVNAEIKGFT LSLHAAAAGK PQLEAQHSAG
1451 IKLGYRW*

```

The following partial DNA sequence was identified in *N. meningitidis* <SEQ ID 88>:

orf46-2.seq

```

1 TTGGGCATT CCCGCAAAT ATCCCTTATT CTGTCCATAC TGGCAGTGTG
51 CCTGCCGATG CATGCACACG CCTCAGATTT GGCAAACGAT TCTTTTATCC
101 GGCAGGTTCT CGACCGTCAG CATTTCGAAC CCGACGGGAA ATACCACCTA
151 TTCGGCAGCA GGGGGGAAC TGCCGAGCGC AGCGGCCATA TCGGATTGGG
201 AAAAATACAA AGCCATCAGT TGGGCAACCT GATGATTCAA CAGGCGGCCA
251 TTAAAGGAAA TATCGGCTAC ATTGTCCGCT TTTCCGATCA CGGGCACGAA
301 GTCCATTCCC CCTTCGACAA CCATGCCTCA CATTCCGATT CTGATGAAGC
351 CCGTAGTCCC GTTGACGGAT TTAGCCTTTA CCGCATCCAT TGGGACGGAT

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401  ACGAACACCA TCCCGCCGAC GGCTATGACG GGCCACAGGG CGGCGGCTAT
451  CCCGCTCCCA AAGGCGCGAG GGATATATAC AGCTACGACA TAAAAGGCGT
501  TGCCCAAAAT ATCCGCCTCA ACCTGACCGA CAACCGCAGC ACCGGACAAC
551  GGCTTGCCGA CCGTTTCCAC AATGCCGGTA GTATGCTGAC GCAAGGAGTA
601  GGCGACGGAT TCAAACGCGC CACCCGATAC AGCCCCGAGC TGGACAGATC
651  GGGCAATGCC GCCGAAGCCT TCAACGGCAC TGCAGATATC GTTAAAAACA
701  TCATCGGCGC GGCAGGAGAA ATTGTCGGCG CAGGCGATGC CGTGCAGGGC
751  ATAAGCGAAG GCTCAAACAT TGCTGTCATG CACGGCTTGG GTCTGCTTTC
801  CACCGAAAAC AAGATGGCGC GCATCAACGA TTTGGCAGAT ATGGCGCAAC
851  TCAAAGACTA TGCCGCAGCA GCCATCCGCG ATTGGGCAGT CCAAAACCCC
901  AATGCCGCAC AAGGCATAGA AGCCGTCAGC AATATCTTTA TGGCAGCCAT
951  CCCCATCAAA GGGATTGGAG CTGTTCCGGG AAAATACGGC TTGGGCGGCA
1001 TCACGGCACA TCCTATCAAG CGGTCGCAGA TGGGCGCGAT CGCATTGCCG
1051 AAAGGGAAAT CCGCCGTCAG CGACAATTTT GCCGATGCGG CATACGCCAA
1101 ATACCCGTCC CCTTACCATT CCCGAAATAT CCGTTCAAAC TTGGAGCAGC
1151 GTTACGGCAA AGAAAACATC ACCTCCTCAA CCGTGCCGCC GTCAAACGGC
1201 AAAAATGTCA AACTGGCAGA CCAACGCCAC CCGAAGACAG GCGTACCGTT
1251 TGACGGTAAA GGGTTTCCGA ATTTTGAGAA GCACGTGAAA TATGATACGA
1301 AGCTCGATAT TCAAGAATTA TCGGGGGGCG GTATACCTAA GGCTAAGCCT
1351 GTGTTTGATG CGAAACCGAG ATGGGAGGTT GATAGGAAGC TTAATAAAAT
1401 GACAACTCGT GAGCAGGTGG AGAAAAATGT TCAGGAAATA AGGAACGGTA
1451 ATATAAACAG TAACTTTAGC CAACATGCTC AACTAGAGAG GGAATTAAT
1501 AAATAAAAT CTGCCGATGA AATTAATTTT GCAGATGGAA TGGGAAAAT
1551 TACCGATAGC ATGAATGACA AGGCTTTTAG TAGGCTTG TG AAATCAGTTA
1601 AAGAGAATGG CTTCAAAAT CCAGTTGTGG AGTACGTTGA AATAAATGGA
1651 AAAGCATATA TCGTAAGAGG AAATAATRGG GTTTTGTCTG CAGAAATACCT
1701 TGGCAGGATA CATGAATTAA AATTTAAAAA AGTTGACTTT CCTGTTCCCTA
1751 ATACTAGTTG GAAAAATCCT ACTGATGTCT TGAATGAATC AGGTAATGTT
1801 AAGAGACCTC GTTATAGGAG TAAATAA

```

This corresponds to the amino acid sequence <SEQ ID 89; ORF orf46-2>:

orf46-2.pep

```

1  LGISRKISLI LSILAVCLPM HAHASDLAND SFIRQVLDRQ HFEPDGKYHL
51  FGSRGELAER SGHIGLGKIQ SHQLGNLMIQ QAAIKGNIGY IVRFSDHGHE
101 VHSPFDNHAS HSDSDEAGSP VDGFSLYRIH WDGYEHHPAD GYDGPQGGGY
151 PAPKGARDIY SYDIKGVAQN IRLNLTDNRS TGQRLADRFH NAGSMLTQGV
201 GDGFKRATRY SPELDRSGNA AEA FNGTADI VKNIIGAAGE IVGAGDAVQG
251 ISEGSNIAVM HGLGLLSTEN KMARINDLAD MAQLKDYAAA AIRDWAQNP
301 NAAQGIEAVS NIFMAAIIPIK GIGAVRGKYG LGGITAHPIK RSQMGAIALP
351 KGKSAVSDNF ADAAYAKYPS PYHSRNIRSN LEQRYGKENI TSSTVPPSNG
401 KNVKLADQRH PKTGVPPFDGK GFPNFEKHVK YDTKLDIQEL SGGGIPKAKP
451 VFDAPRWEV DRKLNKLTTR EQVEKNVQEI RGNINSNFS QHAQLEREIN
501 KLKSADEINF ADGMGKFTDS MNDKA FSR LV KSVKENGFTN PVVEYVEING
551 KAYIVRGNNR VFAAEYLGRI HELKFKKVDF PVPNTSWKNP TDVLNESGNV
601 KRPRYRSK*

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Using the above-described procedures, the following oligonucleotide primers were employed in the polymerase chain reaction (PCR) assay in order to clone the ORFs as indicated:

Oligonucleotides used for PCR

Table 1

- 116 -

ORF	Primer	Sequence	Restriction sites
279	Forward	CGCGGATCCCATATG-TTGCCTGCAATCACGATT <SEQ ID 90>	BamHI-NdeI
	Reverse	CCCGCTCGAG-TTTAGAAGCGGGCGGCAA <SEQ ID 91>	XhoI
519	Forward	CGCGGATCCCATATG-TTCAAATCCTTTGTCGTCA <SEQ ID 92>	BamHI-NdeI
	Reverse	CCCGCTCGAG-TTTGGCGGTTTTGCTGC <SEQ ID 93>	XhoI
576	Forward	CGCGGATCCCATATG-GCCGCCCCCGCATCT <SEQ ID 94>	BamHI-NdeI
	Reverse	CCCGCTCGAG-ATTTACTTTTTTGATGTCGAC <SEQ ID 95>	XhoI
919	Forward	CGCGGATCCCATATG-TGCCAAAGCAAGAGCATC <SEQ ID 96>	BamHI-NdeI
	Reverse	CCCGCTCGAG-CGGGCGGTATTCGGG <SEQ ID 97>	XhoI
121	Forward	CGCGGATCCCATATG-GAAACACAGCTTTACAT <SEQ ID 98>	BamHI-NdeI
	Reverse	CCCGCTCGAG-ATAATAATATCCCGCGCCC <SEQ ID 99>	XhoI
128	Forward	CGCGGATCCCATATG-ACTGACAACGCACT <SEQ ID 100>	BamHI-NdeI
	Reverse	CCCGCTCGAG-GACCGCGTTGTGCGAAA <SEQ ID 101>	XhoI
206	Forward	CGCGGATCCCATATG-AAACACCGCCAACCGA <SEQ ID 102>	BamHI-NdeI
	Reverse	CCCGCTCGAG-TTCTGTAAAAAAGTATGTGC <SEQ ID 103>	XhoI
287	Forward	CCGGAATTCTAGCTAGC-CTTTCAGCCTGCGGG <SEQ ID 104>	EcoRI-NheI
	Reverse	CCCGCTCGAG-ATCCTGCTCTTTTTTGCC <SEQ ID 105>	XhoI
406	Forward	CGCGGATCCCATATG-TGCGGGACACTGACAG <SEQ ID 106>	BamHI-NdeI
	Reverse	CCCGCTCGAG-AGGTTGTCCTTGTCTATG <SEQ ID 107>	XhoI

EXAMPLE 2

Expression of ORF 919

The primer described in Table 1 for ORF 919 was used to locate and clone ORF 919. The predicted gene 919 was cloned in pET vector and expressed in *E. coli*. The product of

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protein expression and purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 919-His fusion protein purification. Mice were immunized with the purified 919-His and sera were used for Western blot (panel B), FACS analysis (panel C), bactericidal assay (panel D), and ELISA assay (panel E). Symbols: M1, molecular weight marker; PP, purified protein, TP, *N. meningitidis* total protein extract; OMV, *N. meningitidis* outer membrane vesicle preparation. Arrows indicate the position of the main recombinant protein product (A) and the *N. meningitidis* immunoreactive band (B). These experiments confirm that 919 is a surface-exposed protein and that it is a useful immunogen. The hydrophilicity plots, antigenic index, and amphipathic regions of ORF 919 are provided in Figure 10. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 919 and the amino acid sequence encoded thereby is provided in Example 1.

EXAMPLE 3

Expression of ORF 279

The primer described in Table 1 for ORF 279 was used to locate and clone ORF 279. The predicted gene 279 was cloned in pGex vector and expressed in *E. coli*. The product of protein expression and purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 279-GST purification. Mice were immunized with the purified 279-GST and sera were used for Western blot analysis (panel B), FACS analysis (panel C), bactericidal assay (panel D), and ELISA assay (panel E). Symbols: M1, molecular weight marker; TP, *N. meningitidis* total protein extract; OMV, *N. meningitidis* outer membrane vesicle preparation. Arrows indicate the position of the main recombinant protein product (A) and the *N. meningitidis* immunoreactive band (B). These experiments confirm that 279 is a surface-exposed protein and that it is a useful immunogen. The hydrophilicity plots, antigenic index, and amphipathic regions of ORF 279 are provided in Figure 11. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 279 and the amino acid sequence encoded thereby is provided in Example 1.

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EXAMPLE 4

Expression of ORF 576

The primer described in Table 1 for ORF 576 was used to locate and clone ORF 576. The predicted gene 576 was cloned in pGex vector and expressed in *E. coli*. The product of protein purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 576-GST fusion protein purification. Mice were immunized with the purified 576-GST and sera were used for Western blot (panel B), FACS analysis (panel C), bactericidal assay (panel D), and ELISA assay (panel E). Symbols: M1, molecular weight marker; TP, *N. meningitidis* total protein extract; OMV, *N. meningitidis* outer membrane vesicle preparation. Arrows indicate the position of the main recombinant protein product (A) and the *N. meningitidis* immunoreactive band (B).. These experiments confirm that ORF 576 is a surface-exposed protein and that it is a useful immunogen. The hydrophilicity plots, antigenic index, and amphipathic regions of ORF 576 are provided in Figure 12. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 576 and the amino acid sequence encoded thereby is provided in Example 1.

EXAMPLE 5

Expression of ORF 519

The primer described in Table 1 for ORF 519 was used to locate and clone ORF 519. The predicted gene 519 was cloned in pET vector and expressed in *E. coli*. The product of protein purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 519-His fusion protein purification. Mice were immunized with the purified 519-His and sera were used for Western blot (panel B), FACS analysis (panel C), bactericidal assay (panel D), and ELISA assay (panel E). Symbols: M1, molecular weight marker; TP, *N. meningitidis* total protein extract; OMV, *N. meningitidis* outer membrane vesicle preparation. Arrows indicate the position of the main recombinant protein product (A) and the *N. meningitidis* immunoreactive band (B). These experiments confirm that 519 is a surface-exposed protein

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and that it is a useful immunogen. The hydrophilicity plots, antigenic index, and amphipatic regions of ORF 519 are provided in Figure 13. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 519 and the amino acid sequence encoded thereby is provided in Example 1.

EXAMPLE 6

Expression of ORF 121

The primer described in Table 1 for ORF 121 was used to locate and clone ORF 121. The predicted gene *121* was cloned in pET vector and expressed in *E. coli*. The product of protein purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 121-His fusion protein purification. Mice were immunized with the purified 121-His and sera were used for Western blot analysis (panel B), FACS analysis (panel C), bactericidal assay (panel D), and ELISA assay (panel E). Results show that 121 is a surface-exposed protein. Symbols: M1, molecular weight marker; TP, *N. meningitidis* total protein extract; OMV, *N. meningitidis* outer membrane vesicle preparation. Arrows indicate the position of the main recombinant protein product (A) and the *N. meningitidis* immunoreactive band (B). These experiments confirm that 121 is a surface-exposed protein and that it is a useful immunogen. The hydrophilicity plots, antigenic index, and amphipatic regions of ORF 121 are provided in Figure 14. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 121 and the amino acid sequence encoded thereby is provided in Example 1.

EXAMPLE 7

Expression of ORF 128

The primer described in Table 1 for ORF 128 was used to locate and clone ORF 128. The predicted gene *128* was cloned in pET vector and expressed in *E. coli*. The product of protein purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 128-His purification. Mice were immunized with the purified 128-His and sera were used for

- 120 -

Western blot analysis (panel B), FACS analysis (panel C), bactericidal assay (panel D) and ELISA assay (panel E). Results show that 128 is a surface-exposed protein. Symbols: M1, molecular weight marker; TP, *N. meningitidis* total protein extract; OMV, *N. meningitidis* outer membrane vesicle preparation. Arrows indicate the position of the main recombinant protein product (A) and the *N. meningitidis* immunoreactive band (B). These experiments confirm that 128 is a surface-exposed protein and that it is a useful immunogen. The hydrophilicity plots, antigenic index, and amphipathic regions of ORF 128 are provided in Figure 15. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 128 and the amino acid sequence encoded thereby is provided in Example 1.

EXAMPLE 8

Expression of ORF 206

The primer described in Table 1 for ORF 206 was used to locate and clone ORF 206. The predicted gene 206 was cloned in pET vector and expressed in *E. coli*. The product of protein purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 206-His purification. Mice were immunized with the purified 206-His and sera were used for Western blot analysis (panel B). It is worth noting that the immunoreactive band in protein extracts from meningococcus is 38 kDa instead of 17 kDa (panel A). To gain information on the nature of this antibody staining we expressed ORF 206 in *E. coli* without the His-tag and including the predicted leader peptide. Western blot analysis on total protein extracts from *E. coli* expressing this native form of the 206 protein showed a reactive band at a position of 38 kDa, as observed in meningococcus. We conclude that the 38 kDa band in panel B) is specific and that anti-206 antibodies, likely recognize a multimeric protein complex. In panel C is shown the FACS analysis, in panel D the bactericidal assay, and in panel E) the ELISA assay. Results show that 206 is a surface-exposed protein. Symbols: M1, molecular weight marker; TP, *N. meningitidis* total protein extract; OMV, *N. meningitidis* outer membrane vesicle preparation. Arrows indicate the position of the main recombinant protein product (A) and the *N. meningitidis* immunoreactive band (B). These experiments confirm that 206 is a surface-exposed protein and that it is a useful immunogen. The hydrophilicity plots,

- 121 -

antigenic index, and amphipatic regions of ORF 519 are provided in Figure 16. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 206 and the amino acid sequence encoded thereby is provided in Example 1.

EXAMPLE 9

Expression of ORF 287

The primer described in Table 1 for ORF 287 was used to locate and clone ORF 287. The predicted gene 287 was cloned in pGex vector and expressed in *E. coli*. The product of protein purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 287-GST fusion protein purification. Mice were immunized with the purified 287-GST and sera were used for FACS analysis (panel B), bactericidal assay (panel C), and ELISA assay (panel D). Results show that 287 is a surface-exposed protein. Symbols: M1, molecular weight marker. Arrow indicates the position of the main recombinant protein product (A). These experiments confirm that 287 is a surface-exposed protein and that it is a useful immunogen. The hydrophilicity plots, antigenic index, and amphipatic regions of ORF 287 are provided in Figure 17. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 287 and the amino acid sequence encoded thereby is provided in Example 1.

EXAMPLE 10

Expression of ORF 406

The primer described in Table 1 for ORF 406 was used to locate and clone ORF 406. The predicted gene 406 was cloned in pET vector and expressed in *E. coli*. The product of protein purification was analyzed by SDS-PAGE. In panel A) is shown the analysis of 406-His fusion protein purification. Mice were immunized with the purified 406-His and sera were used for Western blot analysis (panel B), FACS analysis (panel C), bactericidal assay (panel D), and ELISA assay (panel E). Results show that 406 is a surface-exposed protein. Symbols: M1, molecular weight marker; TP, *N. meningitidis* total protein extract; OMV, *N.*

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meningitidis outer membrane vesicle preparation. Arrows indicate the position of the main recombinant protein product (A) and the *N. meningitidis* immunoreactive band (B). These experiments confirm that 406 is a surface-exposed protein and that it is a useful immunogen. The hydrophilicity plots, antigenic index, and amphipatic regions of ORF 406 are provided in Figure 18. The AMPHI program is used to predict putative T-cell epitopes (Gao et al 1989, *J. Immunol* 143:3007; Roberts et al. 1996, *AIDS Res Human Retroviruses* 12:593; Quakyi et al. 1992, *Scand J Immunol Suppl* 11:9). The nucleic acid sequence of ORF 406 and the amino acid sequence encoded thereby is provided in Example 1.

The foregoing examples are intended to illustrate but not to limit the invention.

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Claims

1. A method for identifying an amino acid sequence, comprising the step of searching for putative open reading frames or protein-coding sequences within one or more of *N. meningitidis* nucleotide sequences selected from the group consisting of SEQ ID NO 1 and the NMB open reading frames.
2. A method according to claim 1, comprising the steps of searching a *N. meningitidis* nucleotide sequence for an initiation codon and searching the upstream sequence for an in-frame termination codon.
3. A method for producing a protein, comprising the step of expressing a protein comprising an amino acid sequence identified according to any one of claims 1-2.
4. A method for identifying a protein in *N. meningitidis*, comprising the steps of producing a protein according to claim 3, producing an antibody which binds to the protein, and determining whether the antibody recognises a protein produced by *N. meningitidis*.
5. Nucleic acid comprising an open reading frame or protein-coding sequence identified by a method according to any one of claims 1-2.
6. A protein obtained by the method of claim 3.
7. Nucleic acid comprising one or more of the *N. meningitidis* nucleotide sequences selected from the group consisting of SEQ ID NO 1 and the NMB open reading frames.
8. Nucleic acid comprising a nucleotide sequence having greater than 50% sequence identity to a nucleotide sequence selected from the group consisting of SEQ ID NO 1 and the NMB open reading frames.

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9. Nucleic acid comprising a fragment of a nucleotide sequence selected from the group consisting of SEQ ID NO 1 and the NMB open reading frames.

10. Nucleic acid according to claim 9, wherein the fragment is unique to the genome of *N. meningitidis*.

11. Nucleic acid complementary to the nucleic acid of any one of claims 7-10.

12. A protein comprising an amino acid sequence encoded within one or more of the *N. meningitidis* nucleotide sequences selected from the group consisting of SEQ ID NO 1 and the NMB open reading frames.

13. A protein comprising an amino acid sequences having greater than 50% sequence identity to an amino acid sequence encoded within one or more of the *N. meningitidis* nucleotide sequences selected from the group consisting of SEQ ID NO 1 and the NMB open reading frames.

14. A protein comprising a fragment of an amino acid sequence encoded within one or more of the *N. meningitidis* nucleotide sequences selected from the group consisting of SEQ ID NO 1 and the NMB open reading frames.

15. Nucleic acid encoding a protein according to any one of claims 6-8.

16. A computer, a computer memory, a computer storage medium or a computer database containing the nucleotide sequence of a nucleic acid according to any one of claims 7-11.

17. A computer, a computer memory, a computer storage medium or a computer database containing one or more of the *N. meningitidis* nucleotide sequences selected from the group consisting of SEQ ID NO 1 and the NMB open reading frames.

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18. A polyclonal or monoclonal antibody which binds to a protein according to any one of claims 12-14 or 6.

19. A nucleic acid probe comprising nucleic acid according to any one of claims 5, 7-10, or 15.

20. An amplification primer comprising nucleic acid according to any one of claims 5, 7-10, or 15.

21. A composition comprising (a) nucleic acid according to any one of claims 5, 7-10, or 15; (b) protein according to any one of claims 12-14; and/or (c) an antibody according to claim 18.

22. The use of a composition according to claim 21 as a medicament or as a diagnostic reagent.

23. The use of a composition according to claim 21 in the manufacture of (a) a medicament for treating or preventing infection due to Neisserial bacteria and/or (b) a diagnostic reagent for detecting the presence of Neisserial bacteria or of antibodies raised against Neisserial bacteria.

24. A method of treating a patient, comprising administering to the patient a therapeutically effective amount of a composition according to claim 21.

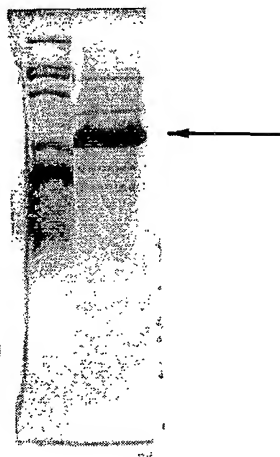
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FIG. 1A

919 (46 kDa)

PURIFICATION

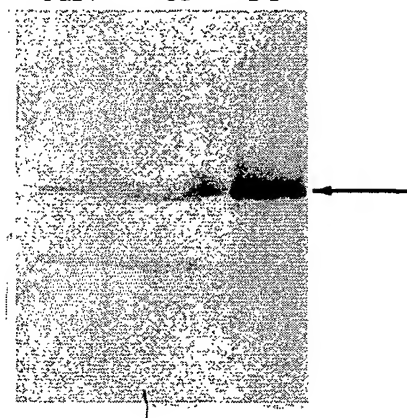
M1 919

*FIG. 1B*

919 (46 kDa)

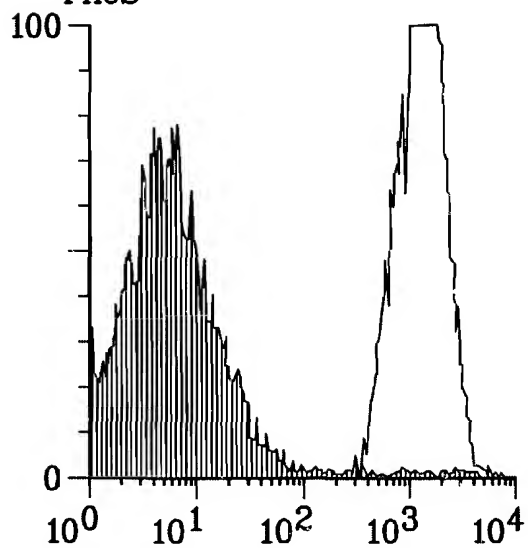
WESTERN BLOT

OMV TP PP

*FIG. 1C*

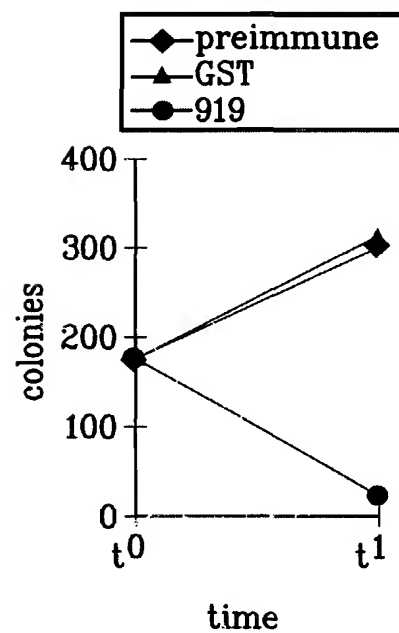
919 (46 kDa)

FACS

*FIG. 1D*

919 (46 kDa)

BACTERICIDAL ASSAY

*FIG. 1E*

919 (46 kDa)

ELISA assay: positive

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FIG. 2A

279 (10.5 kDa)

PURIFICATION

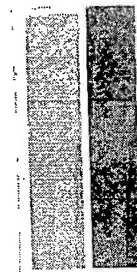
M1 279

*FIG. 2B*

279 (10.5 kDa)

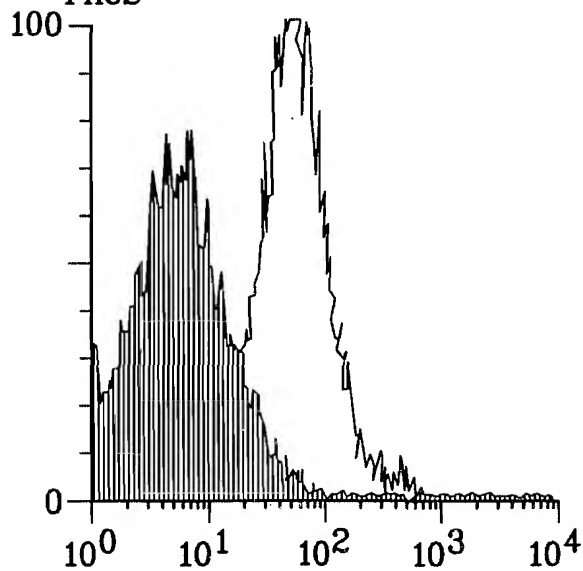
WESTERN BLOT

TP OMV

*FIG. 2C*

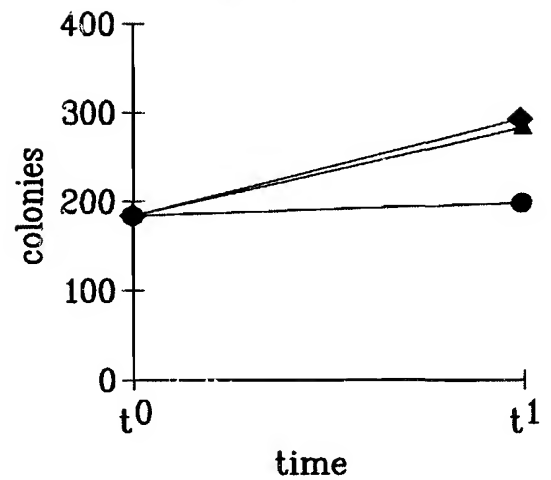
279 (10.5 kDa)

FACS

*FIG. 2D*

279 (10.5 kDa)

BACTERICIDAL ASSAY

*FIG. 2E*

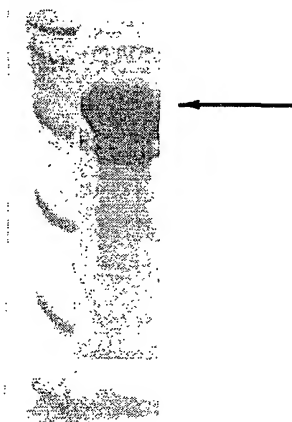
279 (10.5 kDa)

ELISA assay: positive

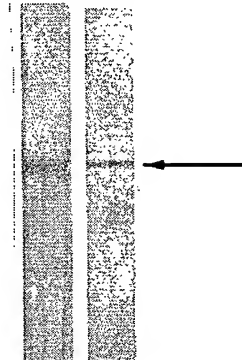
3/18

FIG. 3A

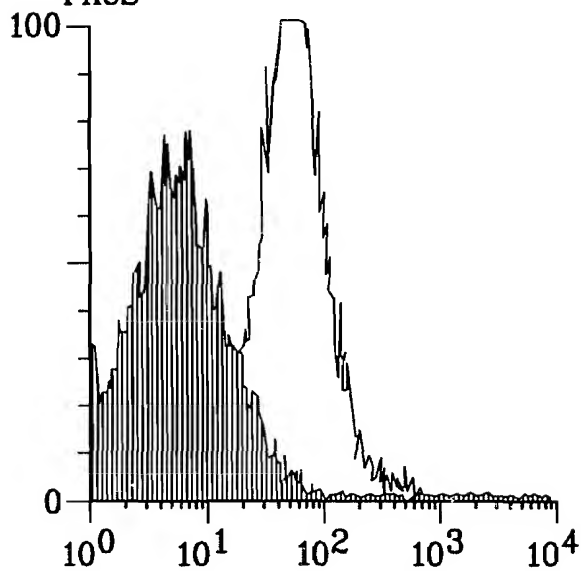
576 (27.8 kDa)
PURIFICATION
M1 576

*FIG. 3B*

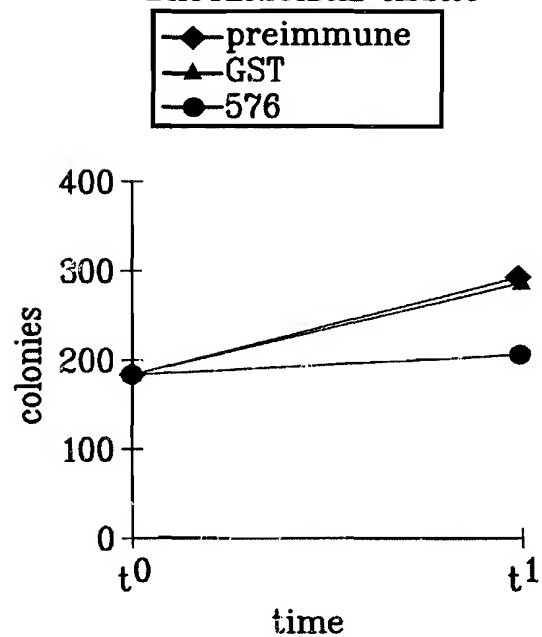
576 (27.8 kDa)
WESTERN BLOT
TP OMV

*FIG. 3C*

576 (27.8 kDa)
FACS

*FIG. 3D*

576 (27.8 kDa)
BACTERICIDAL ASSAY

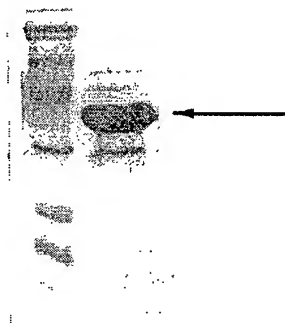
*FIG. 3E*

576 (27.8 kDa)
ELISA assay: positive

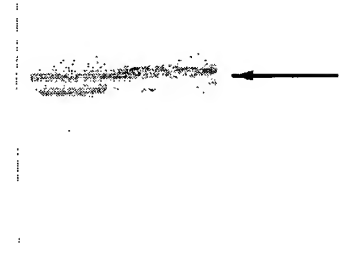
4/18

FIG. 4A

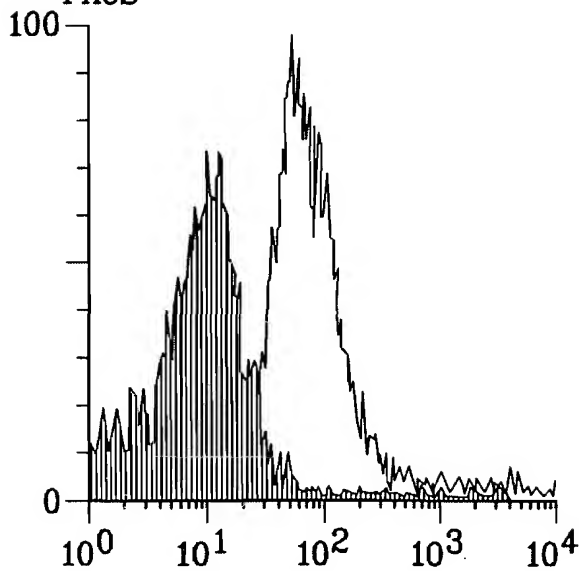
519 (33 kDa)
PURIFICATION
M1 519

*FIG. 4B*

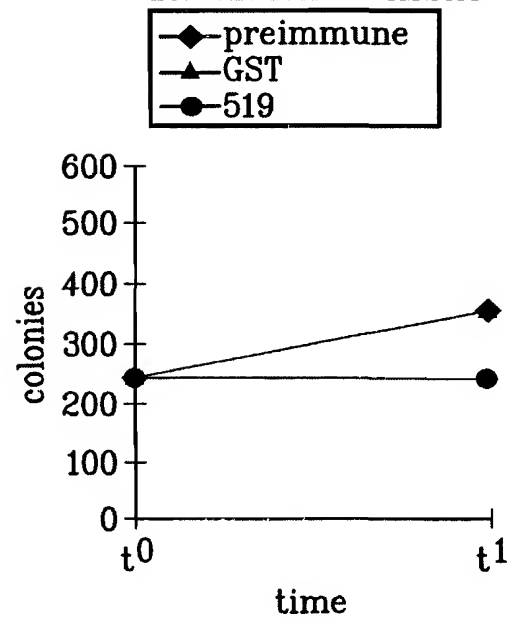
519 (33 kDa)
WESTERN BLOT
TP OMV

*FIG. 4C*

519 (33 kDa)
FACS

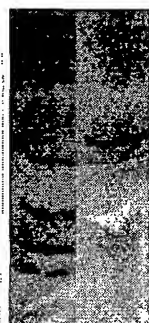
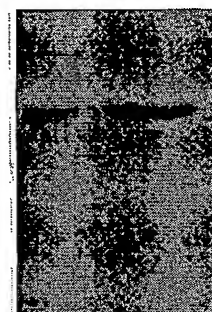
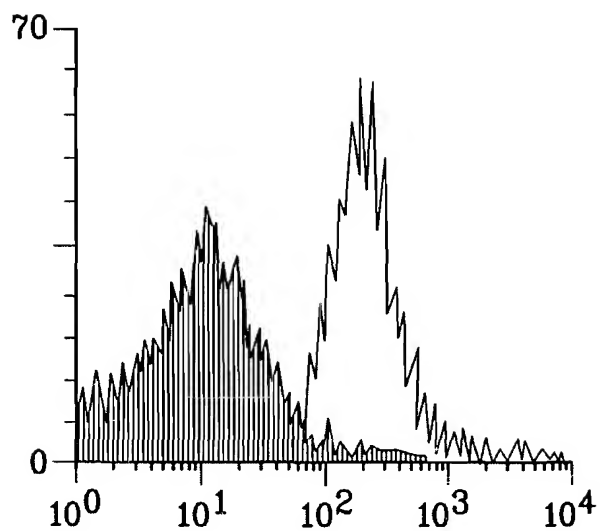
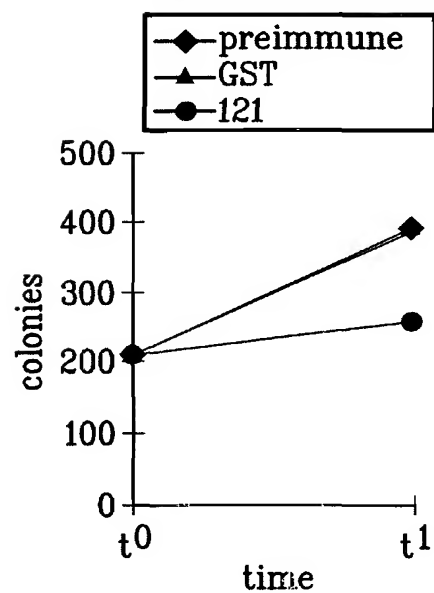
*FIG. 4D*

519 (33 kDa)
BACTERICIDAL ASSAY

*FIG. 4E*

519 (33 kDa)
ELISA assay: positive

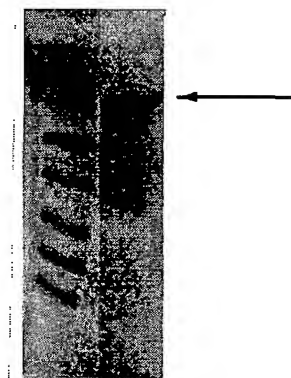
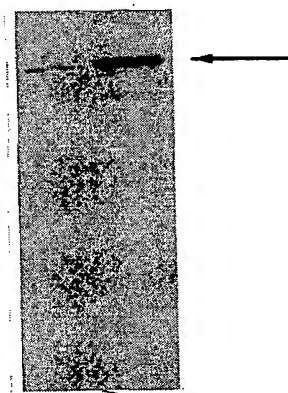
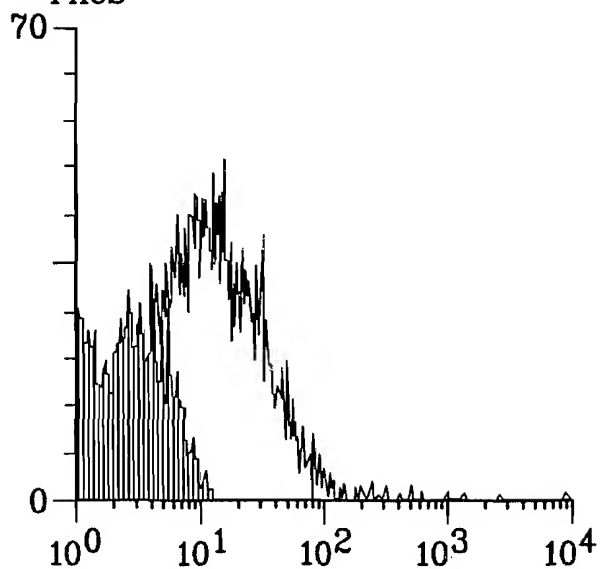
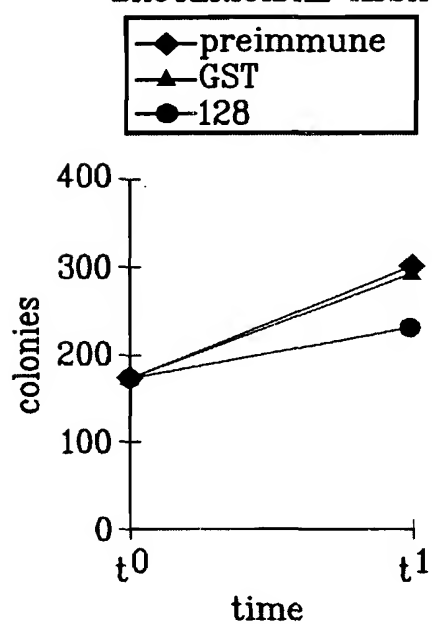
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*FIG. 5A*121 (40 kDa)
PURIFICATION
M1 121*FIG. 5B*121 (40 kDa)
WESTERN BLOT
TP OMV*FIG. 5C*121 (40 kDa)
FACS*FIG. 5D*121 (40 kDa)
BACTERICIDAL ASSAY*FIG. 5E*

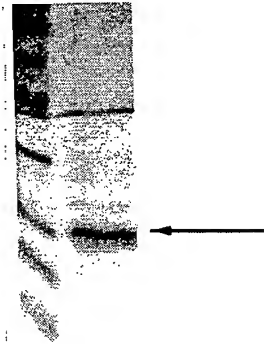
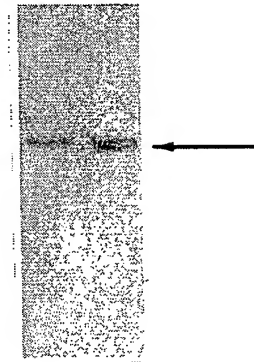
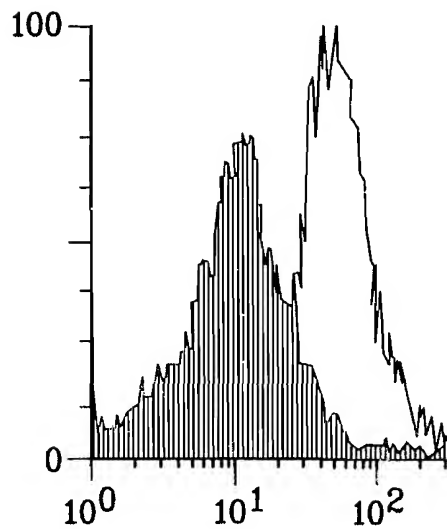
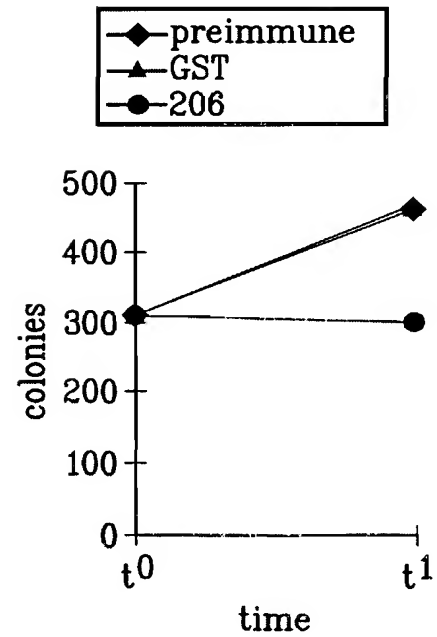
121 (40 kDa)

ELISA assay: positive

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*FIG. 6A*128 (101 kDa)
PURIFICATION
M1 128*FIG. 6B*128 (101 kDa)
WESTERN BLOT
TP OMV*FIG. 6C*128 (101 kDa)
FACS*FIG. 6D*128 (101 kDa)
BACTERICIDAL ASSAY*FIG. 6E*128 (101 kDa)
ELISA assay: positive

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*FIG. 7A*206 (17 kDa)
PURIFICATION
M1 206*FIG. 7B*206 (17 kDa)
WESTERN BLOT
TP OMV*FIG. 7C*206 (17 kDa)
FACS*FIG. 7D*206 (17 kDa)
BACTERICIDAL ASSAY*FIG. 7E*

206 (17 kDa)

ELISA assay: positive

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FIG. 8A

287 (78 kDa)
PURIFICATION
M1 287

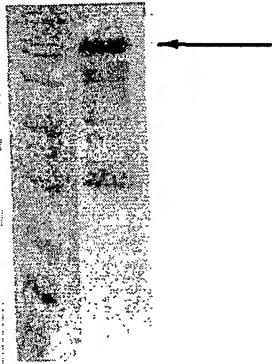


FIG. 8B

287 (78 kDa)
FACS

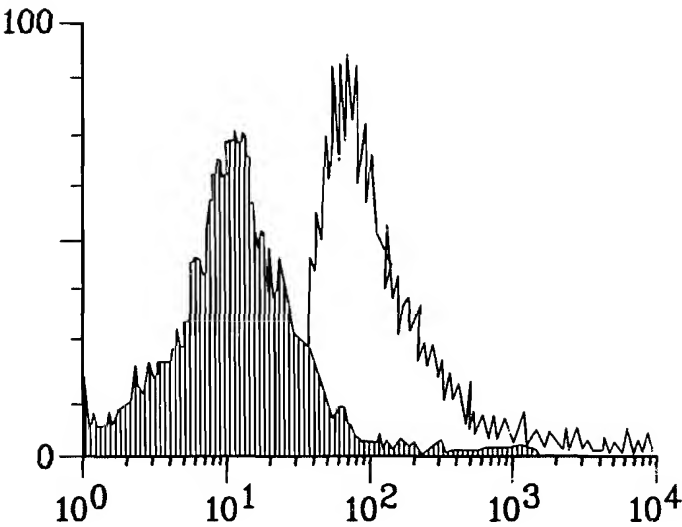


FIG. 8C

287 (78 kDa)
BACTERICIDAL ASSAY

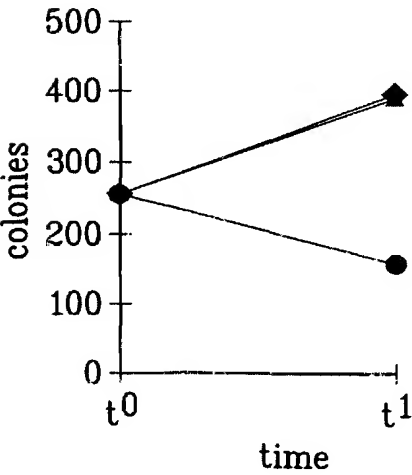
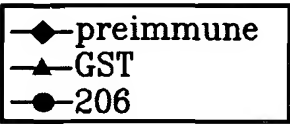
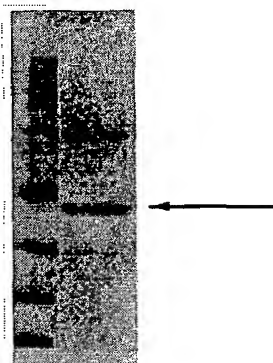
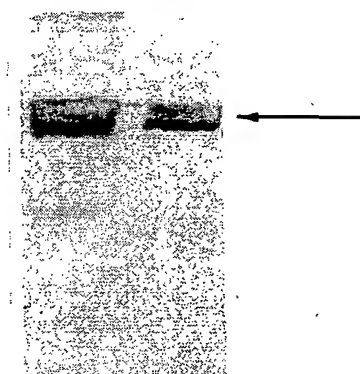
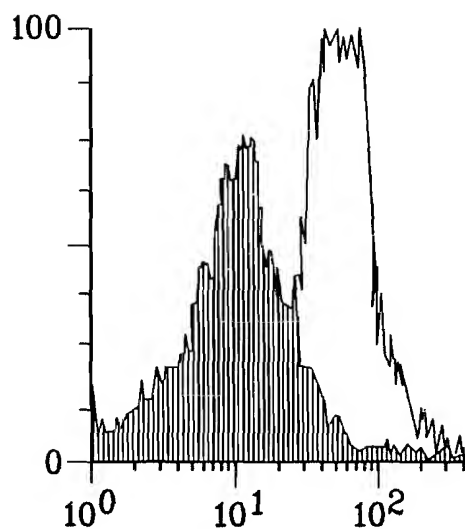
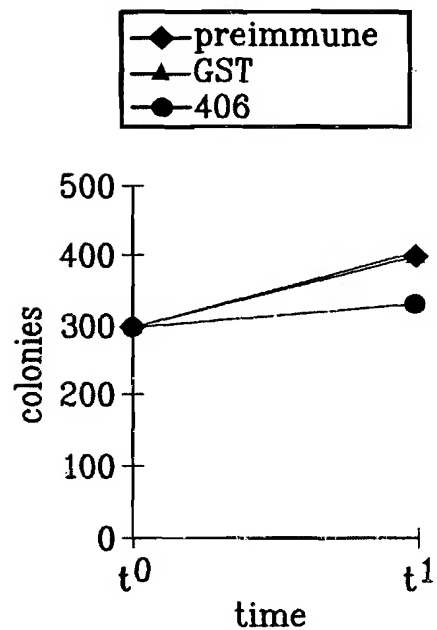


FIG. 8D

287 (78 kDa)
ELISA assay: positive

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*FIG. 9A*406 (33 kDa)
PURIFICATION
M1 406*FIG. 9B*406 (33 kDa)
WESTERN BLOT
TP OMV*FIG. 9C*406 (33 kDa)
FACS*FIG. 9D*406 (33 kDa)
BACTERICIDAL ASSAY*FIG. 9E*406 (33 kDa)
ELISA assay: positive

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919

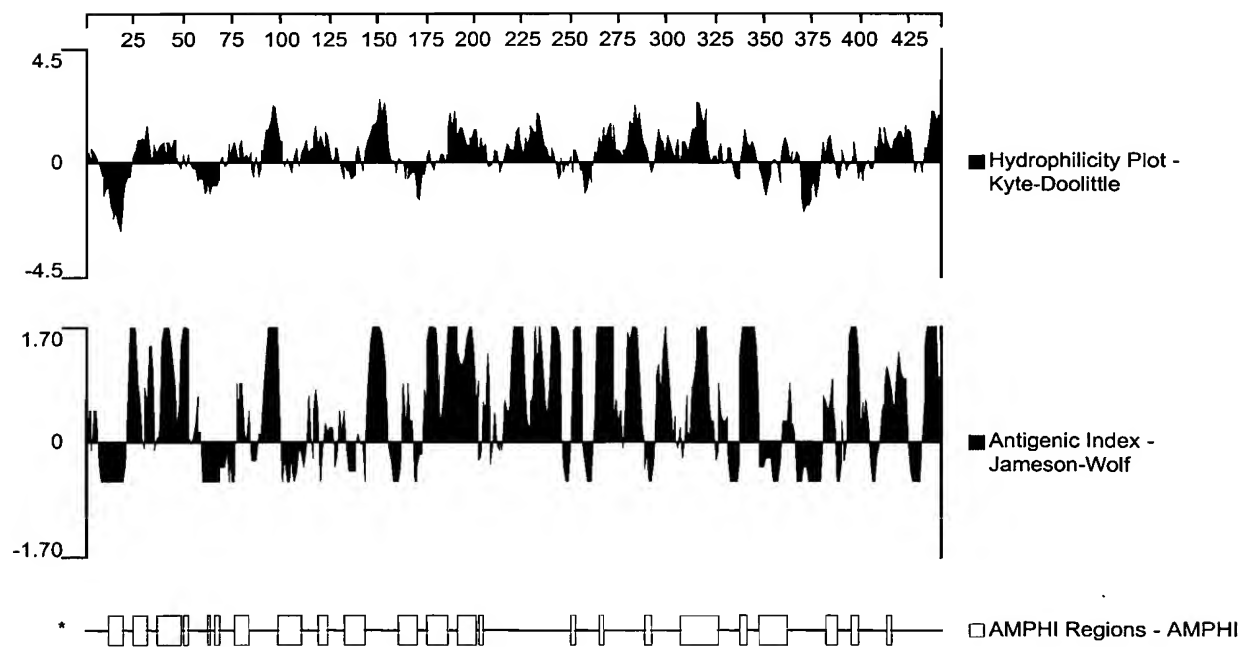
Hydrophilicity Plot, Antigenic Index and AMPHI Regions

Fig. 10

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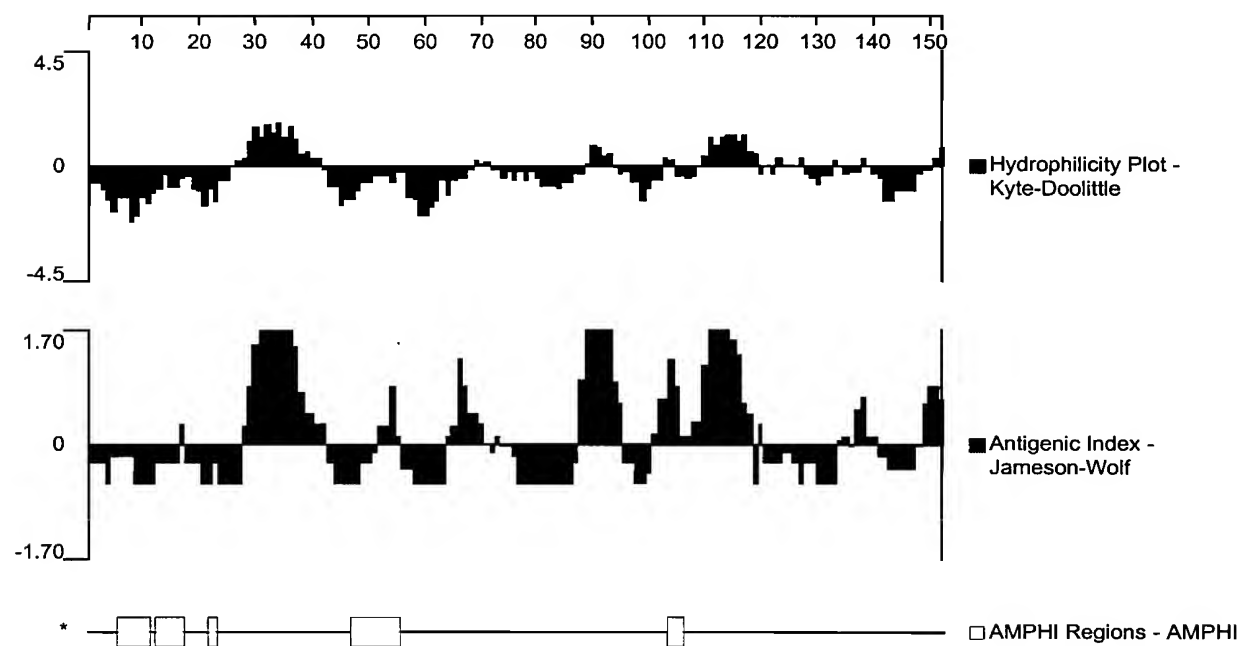
279**Hydrophilicity Plot, Antigenic Index and AMPHI Regions**

Fig. 11

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576-1
Hydrophilicity Plot, Antigenic Index and AMPHI Regions

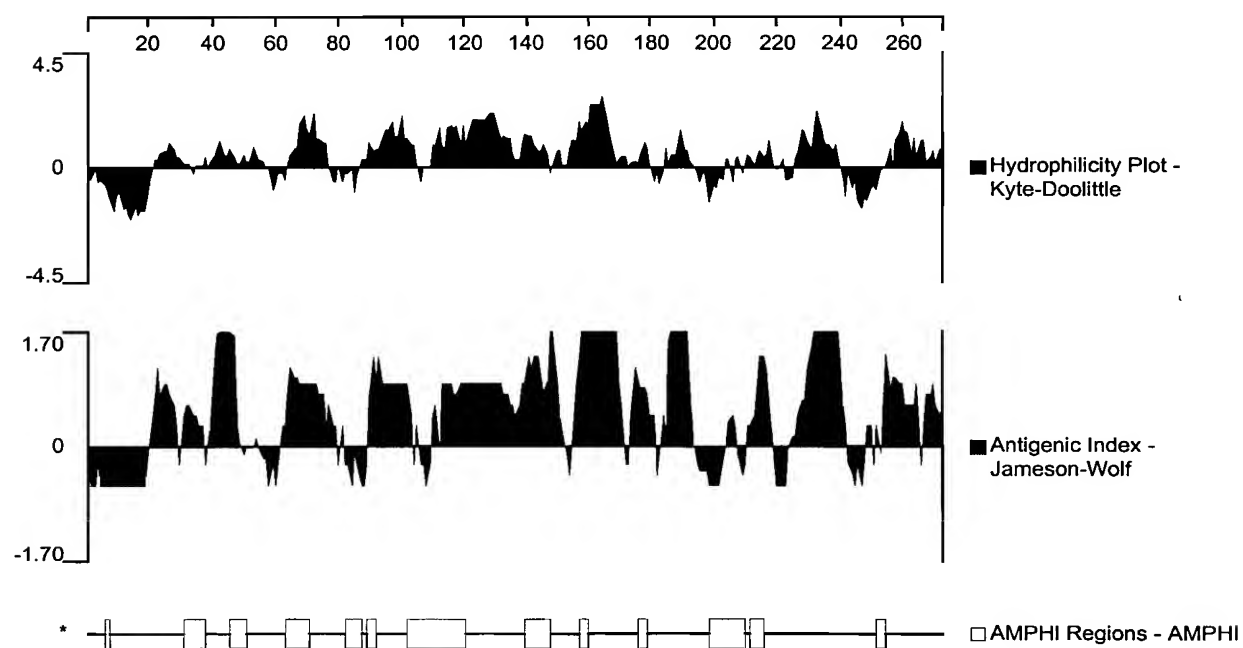


Fig. 12

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519-1
Hydrophilicity Plot, Antigenic Index and AMPHI Regions

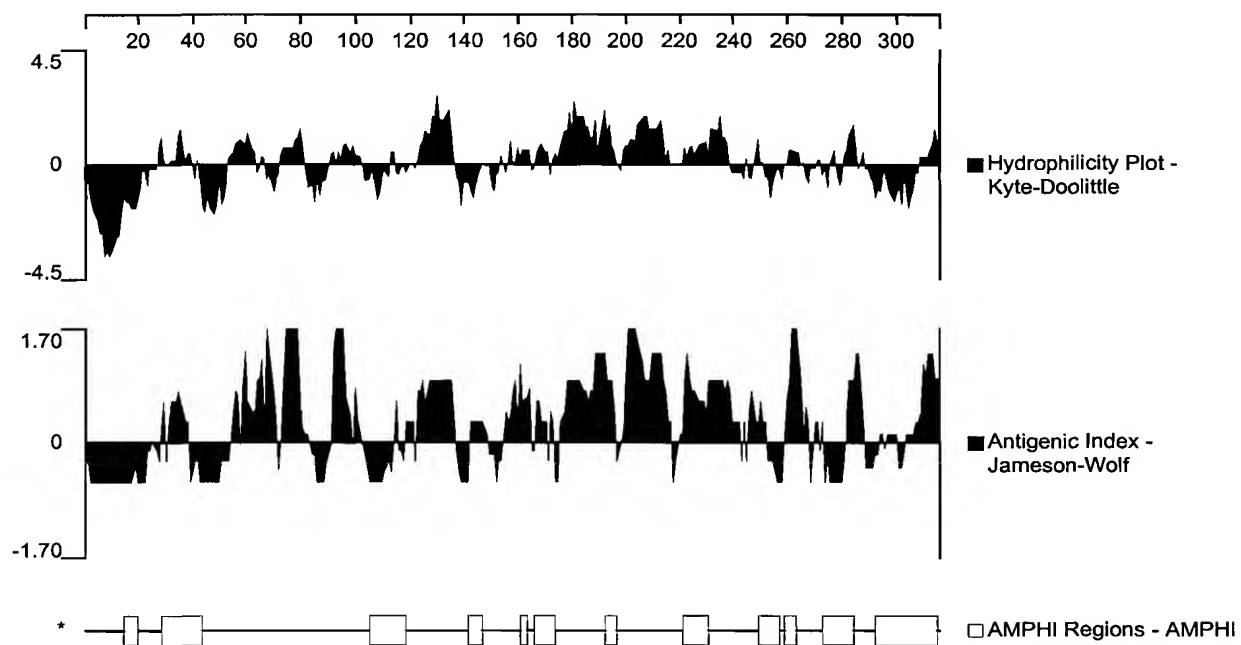


Fig. 13

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121-1
Hydrophilicity Plot, Antigenic Index and AMPHI Regions

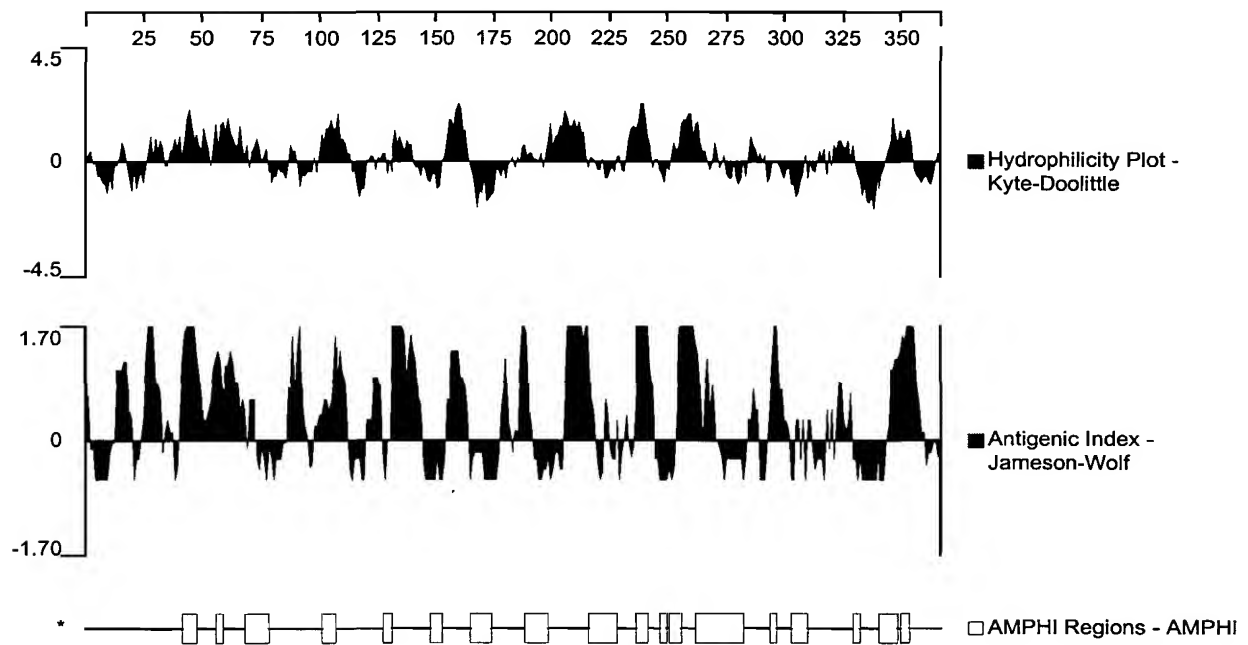


Fig. 14

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128-1
Hydrophilicity Plot, Antigenic Index and AMPHI Regions

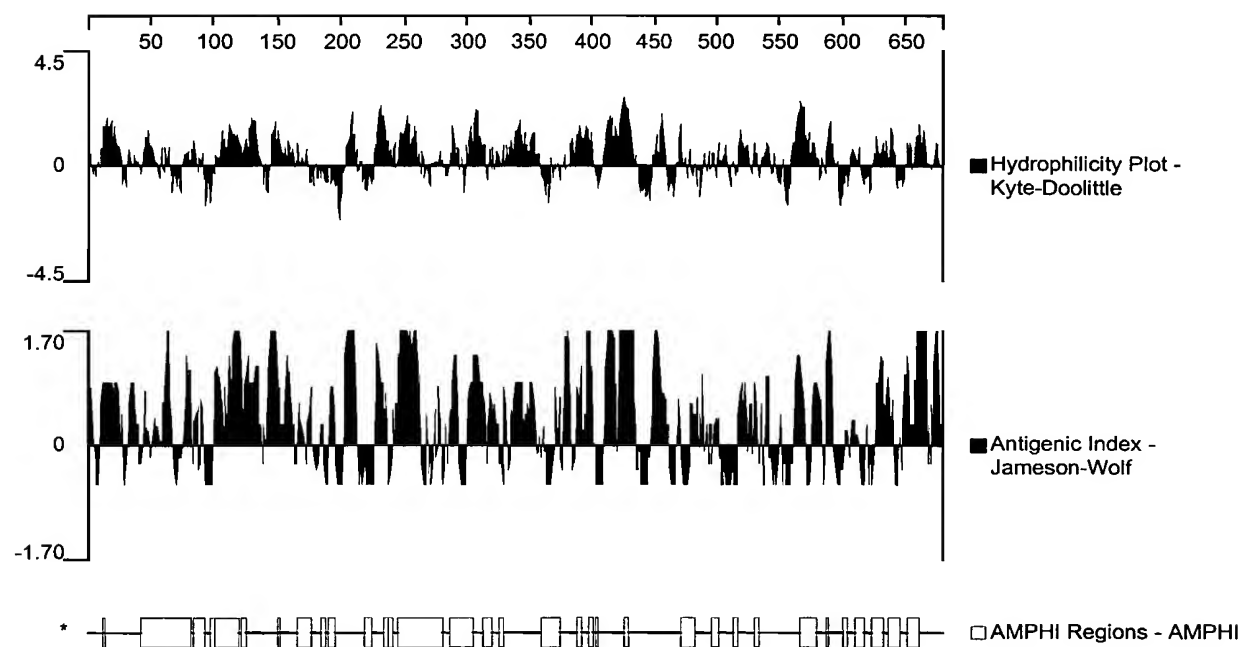


Fig. 15

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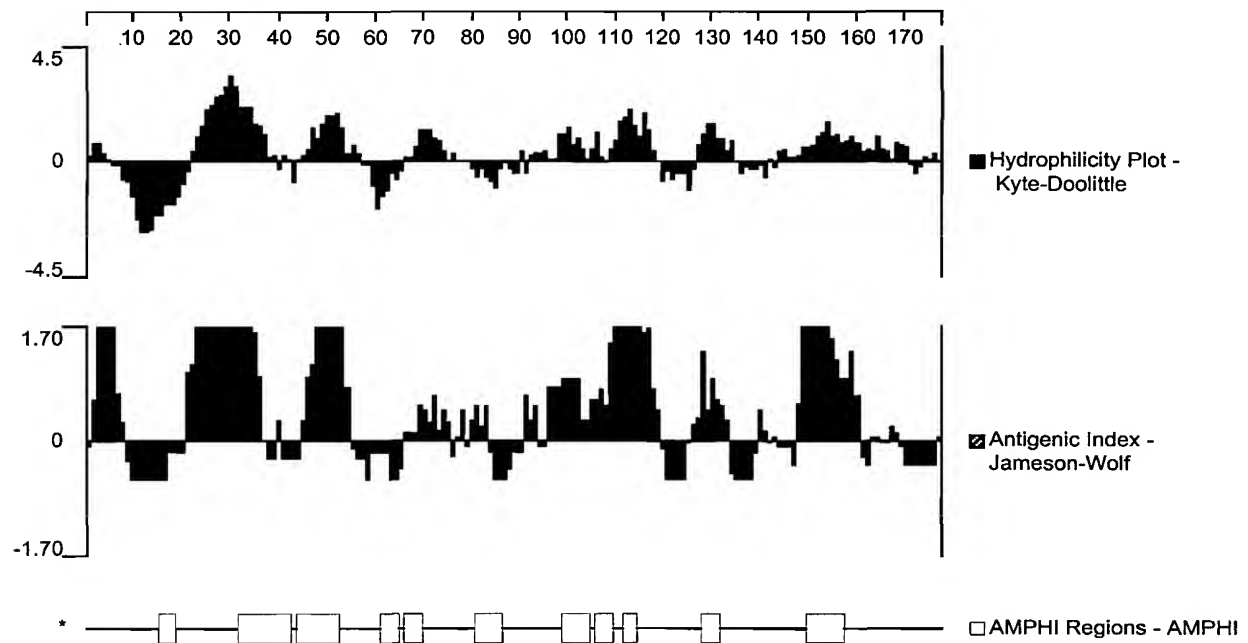
206Hydrophilicity Plot, Antigenic Index and AMPHI Regions

Fig. 16

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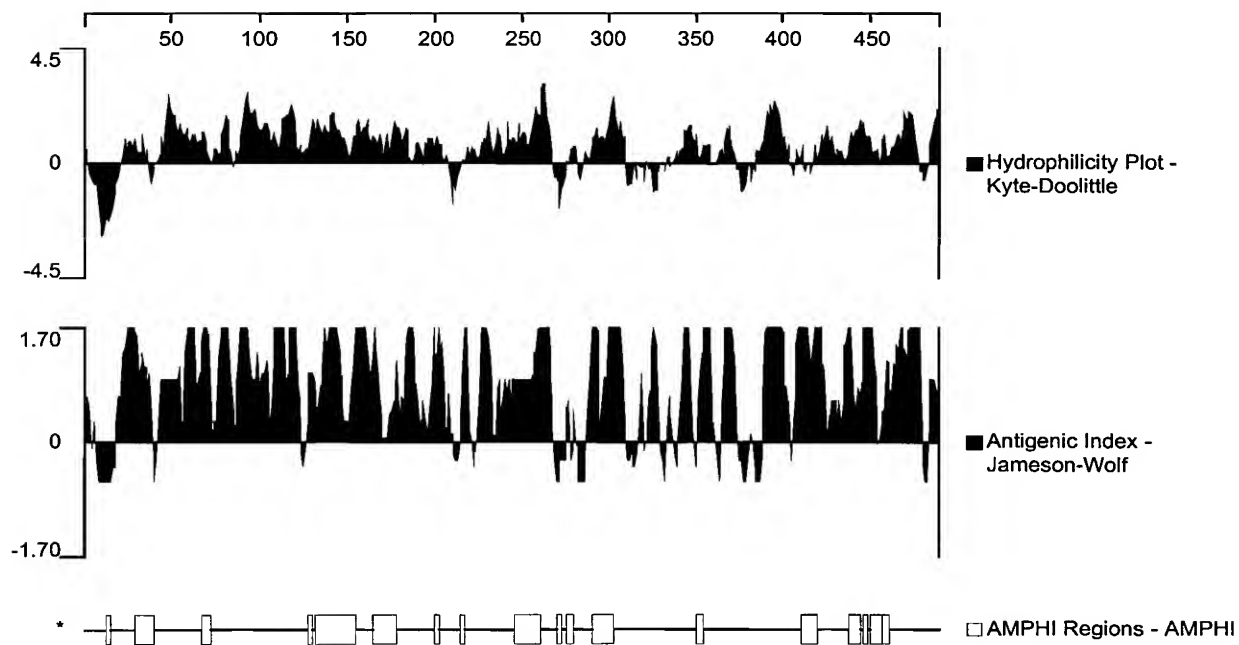
287Hydrophilicity Plot, Antigenic Index and AMPHI Regions

Fig. 17

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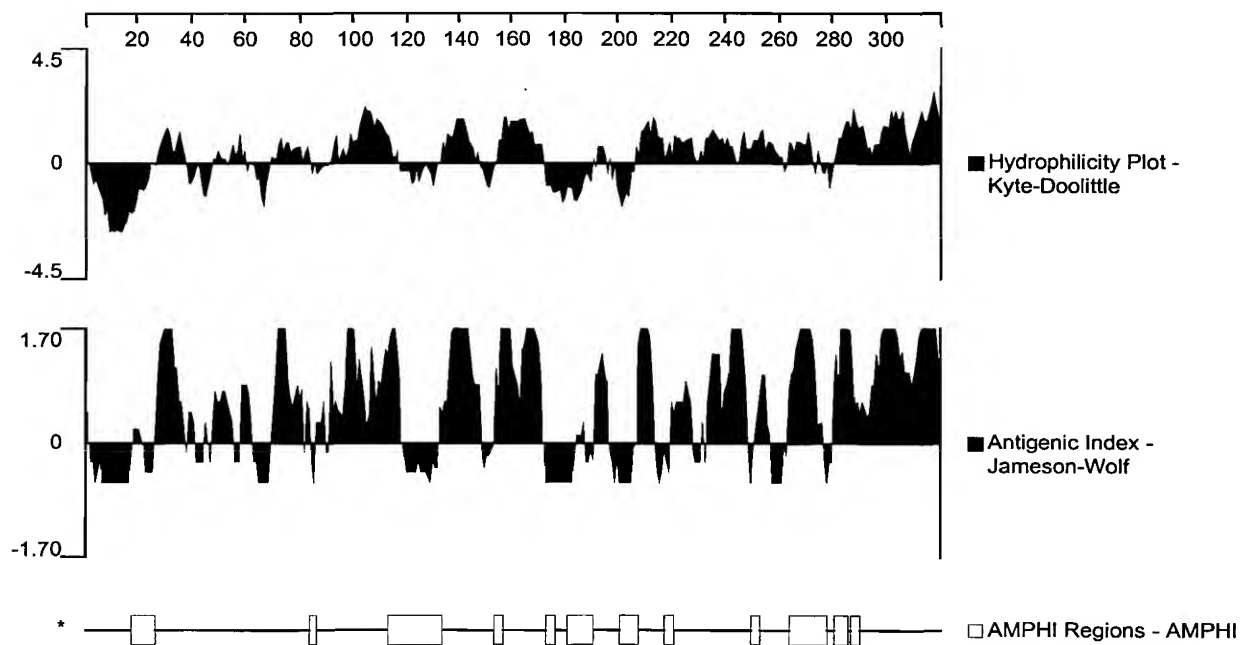
406**Hydrophilicity Plot, Antigenic Index and AMPHI Regions**

Fig. 18

Appendix A

-1-

APPENDIX A

The following DNA sequence was identified in *N. meningitidis B* <SEQ ID NO. 1>:

TAAACCTTATCCACATCCAAACGCATAACCGTAACCCATTACCGTTATGGAATGTGCG
CCGACAACCACCCAGCCGAATGATTCATAAAATATTTGCACATCAGGCGTATAAAGATAC
AAGAACCTTATCCCCAGCGAACGCGCTGCGCCTATGCAGTGGGCGACAGCCTCCTGCCA
ATGCCTTTTCCGCGATATTCAGGTAAACAAAGACATCCCCAACCAATATTCATACCGT
GGAAACTTTCCATATCATGCCGCTTGACCGCAGCCGAACCAACAGGATTCGGAATCA
TCCACAGCCGCAATGCCAGCGGCAGTTTCGTATCCTTCAAACACCTGCCGTAATAGGCA
TGAATCTTATCCACAGAAGACCACGGTTCAAATCCGTGCCACTCCTCAAACAACGCCTGA
ACCAACCTGCCGATATGCCCGGCTTTCAGCCGTGTAATGAAAACAGTATTTGTCCACAAAG
AGGGAATTCATCGGTCAATTTCCCGACGCTTCGTTCCCGCTGCCCGTAAACCGCATTC
CAAGCATGGTCCAAACGCATCCGATTTGCCCTCAAATCTTCAGCCTGCCGGGCTTTTTCG
GCCATTGCTGCAGGAATTTCCGCTTCCAAACGGGCGATGTCTGCCTGAGCCGTCTGCAAA
CGCCGGCGCGCATCTTCCAAATCCGACTGCATCCCGATGATTTTCCGTCCAGATTGTTT
TGCTTTTGAATAAGCGCGGTAAACCGGATTGGATGCTGAGCAGATTGTCTTCAGCATCC
CCTGCCCATACGCTTGTAGAAAAAACAACCATCAGAAAAATAAAATATTTTTTTCATTTTT
AATTTCCATTTAATGCTGTCTGAAGCCGATTCGACATCAGACGGCATCGCCACGCGC
TGTGGATAACTTAAGCGCGGATGCGTTTCAACACTTCTTCTTGGCGATTATGCCAACA
CAGCATCCGACGCTGGGGGTTTTCGCGGTACCGCAGACGGCAAGGCGCAGGGCATGCCGA
GTTTGGCCATTTTAAATGCTTCTTCGTCGCAGAAGGGTTTGAAGAGTCTGTGATGGCTT
CGGCATTCCAGTCTTCCAGCCCTTCGAGGCGTTTCGGCAAAGCGCAGCATACGGGCGGCGG
CTTCATCGTCCCAAGTGTTCCTGCACGCTCTGCTTCGGCAGGCGTTTGTTCGACGTAGAAGT
AGAAGCACTCGTCGGCAAGCGTGTCAAGTCTTGGGGCGGCTTTTGACCACTTCCAACA
CATCTTCCAAAGCAGGTTTTTCGGTTTCATGAATATCGCGCAACGCAAGGCGGGGTTTGA
CGAGTTCCGGCAGTTTGGCGTTGGGTGTGATTTTGATGTGTTTCGCGCTGATCCAGTAGA
GTTTTTCAAGTCCATACGGCTTGGAGACGGGGAACGCTTTTCAAATCAAACCATTCGA
TGAATTGTTCATTTGTGAAGAATTCATCGTCCCGTGGCGCCAGCCCAAGCGTCCAGAT
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CGCCGCTCGCTTTGGAGATTTTTTTTCGCTTGTTCGTTAAGAATCATCGCCAGGTGGCCGT
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Appendix A

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Appendix A

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Appendix A

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Appendix A

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Appendix A

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Appendix A

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Appendix A

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Appendix A

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Appendix A

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CGTACTGCTAAGGGTGTACGTGCTGCTGATGGTGCCTTAATTAATTCGATAACAATGCC
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CGTGAATTGCGTACTGAGCGATTATGAAAATCGTTTCATTGGCACCTGAAAGTATTATAA
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ATTATCCACTCCATGCGAGGAGAACTGGTTTCTGTGGCGGGTATTTACCGTAATTTTGA
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TCTTCTACGCTTTCGGTACGCGTGGCTTTGGCTGTGCGGTGTCGATAAAGGCTTTGAGC
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GCGGCTTTGGCAAGGGCGATGGCGATTTTGCTTTGAGGTGGTGGGCGAGTGTGTGTCC
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TGTCGGCTGCTAAGCGTTTGGCGATGCCGCGCGGTAGGCGCGGTAGGCTTTGAGGGTT
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[illegible]

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TTCGCGTACTGCCAGCGGTATCTGCTGCGCTGACCAAGGGACATACCGCAATGAGCG
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ACCATTGACTGCAATCCCTGCCGGATTGGCAATAACGACTTCTGCACGTCGTCCGCCAC
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GGCCAAACAAGAGTTGGTTGTACCTAAGTCGATACCGATTACCTTTGCCATGTGGATAAT
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GGACGATCTGCTGCAAGTTGCGCGAGGCAATGGAAGGCAACGTGCGCGAGCGTTATGAGCT
GCTTTCAAGCGCGGAGATCGTCCGAAATTCGTGTATCAGCAACAGGTCGAGCTGAC
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AGCCAATGATACGCTTTTGTGCTGTCGCGGTTCTCTTCCCGCACCGATGCAGATTGT
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GCGGACGAGGCAATCAATCTCGGTTTCAACACGATGGGCGACCATTTGTCGATTTCGAA
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TTTTGCGGTGCTACACTGTTTGGGTATTCTGACTTCGATGTATTTCATCCGTCGTCGTATT
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TGCGTTCAAGCCGCTTTTGCAAAGGCCGCGCTTTCGGCAAGCGGACACGGACACTGCCGA
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GCAGGGATTTCGGTAAACCGCGCTAATAGCCGGTGTGCCACAGGTACAGGGCGGCGGCAA
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TCGGTCAAGCTTTTGGGTTTTACTGCCCAAAACCTTATGGACAATCAAACCGAAACAAATG
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TCGCCGGCAAGCATCAGGAAGATGGCGGGAGTCAGCAATGGGGAAATCAGGGTGGAAACA
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ATCGGTACAGGCCGTTTGGCTAAACGTGCATTGTTGGCCGTATTGCCGAAACCTGAAGAT
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GCCGCCGATTTGGAGCCCTTCAGACGGCATTTGGTTCGATCAGCCAGCGCGCGGAGTTT
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GGCTTTTCTGCGGTGACGACGGGGTTTTTAAAGAACTGCCGACATTGCCAAGCACGTT
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Appendix A

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CACCCGCGCCGGTATGCTGACCAATCCGTTGCGCGACCGCTTCGGCATCGTCTCCCGCCT
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ACGGCAAAACCATATCCCAACCATCAGGAATCCCCCTTATGAACAGACAAAAAGTCAT
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ATTGGGATACGATTATCTCGATACCGGCGCACTCTACCGCTGACTGCCCTATATGCACA
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CGCCGTATTTTCAGGCAGCCGCATCCTGCTCGGCGGCGAAGACGTTTCAGACGGCATCCG
GACAGAAAGCCATCGGCATGGGCGCATCCGCGAGTCGCACAGTTGCCTAAAGTCCGCGCCGC
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CACC GGATCGGTCGCTCTTCCCCCAAGCCGAACCTTAAATCTTCTGACGGCAGAATCCAA
AATCCGTGCCGAACGCCGCCGCCAAACAAATCGGCATCCCTGCGAAGGTTTGGCATTCTGA
GCGCATCTGTCCGACATCGAAGCCAGAGACGAGGCGAGACC GAAACCCGCAAAGTTGCCCC
CCTGAAACAACAGCCCGATGCCCTGCTTTTGGACACAAGCCGCTGACTATAGAAGAAAC
TGTA AAAAAAGTGCTTGATTGGTATCGTGAAGTTTAAATTTTCAGGTATAATCGCACAAA
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GCTCAAGGCGAAATTAAGATTAAAGTTCGGCGACTTCGTTACCGTTACCATCGAATCCGTC
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ATTGCCCTGGAAGAACCTGGA A A A C G G C G A C A T C C T G T C C G G C A T C A T C A A C G G A A A A
GTCA A A G G C G C C T G A C C C T A T G A T T A G C A G C A T C C G C G C A T T C C T G C C G G G T C T T T G
G T C G A C G T A C G T C T G T A A A A G A C A C T T C T C A C T T C G A A G G C A A A G A G A T C G A A T T C A A A
G T G A T C A A A C T G G A C A A A A A C G C A A C A C G T C G T T G T T C C C G C C G C G C G T C T G G A A
G C C A C T T T G G G T G A A G A A C G C A A A G C C C T G C T G G A A A A C C T G C A A G A A G G C T C C G T C A T C
A A A G G C A T C G T T A A A A A C A T T A C C G A T T A C G G T G C A T T C G T T G A C T T G G C C G C A T C G A C
G G T C T G T G C A C A T C A C C G A T T T G G C A T G G C G G C G C G T G A A A C A C C G A G T G A A G T C T T G
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T C C T T G G G T A T G A A C A A C T G G G C G A A G A T C C T T G G A G C G G T C T G A C C C G C G T A T C C T
C A A G G C A C C C G C C T G T T C G G C A A A G T A T C C A A C C T G A C C G A C T A C G G C G C A T T C G T C G A A
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G T A C A C C G A G C A A A G T C G T A C A A C T G G G C G A C G A A G T C G A A G T C A T G A T T T T G G A A A T C
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C T G T C C T G G A C C A A T C C G C G A A G A A G C C G T A C G C A A A T A C A A A A A A G G C G A A G A A G T C
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C T G G A A G G C G A T C C G T T C G G C A A C T T C A T C A G C G T G A A C G A C A A A G G T T C T T T G G T T A A A
G G T T C C G T G A A A T C T G T T A C G C C A A A G C T G C T G T T A T C G C C C T G T C G A C G A A G T A G A A
G G C T A C C T G C C T G C T T C C G A A T T T G C A G C C G A C C G C G T T G A A G A T T T G A C C A C C A A A C G
A A A G A A G G C G A C G A A G T T G A A G C C G T C A T C G T T A C C G T T G A C C G C A A A A A C C G A G C A T C
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A C A G G T C G T T T G G A A A G G T T C A G C A T T T G C C G T G T G A A G G T C G C T C T G A A C T G C C G

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TGACGTGTACGGAACAAAATTTTCATTACCAAGTCAGGCTTTACGCTTATGCGGCAGAAC
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GGTGAAGAAACGACACCCGCATTTTCCAGTGCCTCCATCAGGTTTGGCCGCGCGTTATA
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Appendix A

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GCAAC

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CATCGGTGTTCAACACTTCGATGGTCAGCCCTTTCATACTTTTCACGACATCGCCCGGT
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TTGGGGCGCTCCGACCGTTTCCAAAAAGGAATCCAGCTTGTCTGTCGAGCCGGTAATTTCAATCGTATAGCTGCGGTTCGGTTACGTCGATGATGCTGCCCCGGTAGATTTCGGTCAAGCGTAAAAATTCGTCGCGGTCTTTGCCGGCGGCACGGACTTTTACCAACATCAGTTCGCGTTTGACAAAACGGCTTTTCATTCAAAATCGACCACTTAAATCACTTCAATCAATTTATTGAGTTGCTTGGTAATTTGTTTCGATGACCTGCTCGTCGCGGTGGGTAACGATGGTCATCCGTGACAGGGTTTTGTCTTCGGTCGGCGCAACCGCCAAAGAATCGATATTGTAATCCGGTGCAGAGAACAAACCGACACCGCGCTCATCGCACCTGATTCGTTTTCAATCAGAACAGATAAGATATGTCGCATTTGTCTCTCTTACGCCCTTTCCGTCCGCACGCATATGCGGCGGAAGTACCATTTCTGCCAAACCTTTGCCGTTGCCGACCATGGGCATCATTCTGTTTCTGGTCCGTGACGGAAGTCGATAAACACACGCTGTCTTTTGGTTCAATGCTTCCAACAACGCACCTTCCACATCAGACTTCTTGTCCACGCGGATACCGATATGGCCGTATGCCTCGGCAAGTTTGACGAAATCGGGCAAAGAATCGAAATAGGTTTCCGACTCTCGTCCGCCGTAATATATTTCCTGCCACTGGCGTACCATACCGAGATAACCGTTGTTACGCGTAATGACGTTAACCGGAATCCGATATTGGAAACAGCTGGACAGCTCTTGGATGTTCACTCGGATCGAGCCGTCCCGGGTGATACAGATACGCTTGTGATCCGGGGCGGCAAGTTTGCACCAATCGCATAAGGCAGACCCACGCCCACTGTACCCAAACCGCCGGAATTGAGCCATTGGCGCGGACGTTTCAAGGGATAATATTGAGCCGCAACATTTGATGCTGCCCTACATCCGATGTGATGATTGCCGAATTGCCGGTAATCTCGGCAAGCTTCTGAATCACATATTGTGGCTTGATAATTTGCTGCCGTTGTCAAACCACAAGCAATCTCGGGAACGCCATTCTCTATGGTTTCCACCATTTGCCCAAAGCATCTTCAGACGGCACGGACTCTTGTTTTGGCACAGCGCAACCATCTCGGACAAAACGTTTTCACGTCGCCGACAAATCGGAATGCCACCTTACGCGTTTGGCGATGCTGGAAGGATCGACATCGATATGGATAACCTTCTTCGCCCTCTCGAAAAATTTGGACGGTACGGAAACCACACGGTCTGTCAAACCGCGCACCTTACGGCAAGAACGACATCCGCATTCTGCATGGCAAGGTTTGCCCTCGTAAGTACCGTGCATACCGGATACCGAGGAATGGCGGTCCGCCGAAGGATAAGCGCCCAAGCCCATCAGCGTACCGGTGCACGGAGCACCGGTCAATTCGGACAAATCGGGTCAGCTCTTCAGAAGCATTACCCAACACACCGCCGCCCAAATAGACGACCGGACGTTTGGCAGATGCCAACATCTGCACGGCTTTTAACTCTGACCGATATGCTTGAACAACCGGTTGATACGAAGGATAAAAAATGTCTTCTCGAGGATAGCTGAATTTGCCATCGCCGTGCCTGAACATCTTTCGGGACATCAACCACACCGGCCCGGTCGGCCGCTTGCGGCAATTTGGAACGCCTTTTAAATGGTTTCCGCCAACTCATGTATGCTCCGTAACCAGGAATTTGTGTTGACGCACGGACGGTAATACCCACCGTATCAACTTCTTGAACGCATCCGTACCAATCAGGGAATGCGCTACCTGCCCGCTGATGACCACCATCGGAATCGAATCCGTATAGGCAGTAGCAATACCGGTCAGTGCATTGGTAACGCCCGGGCGGATGTAACCAATGCCACGCCACCTTACCGCTGACGCGCGCATACGCATCGCCCGGTACTGCGCGCTGCTCATGGCGGTAAGAATGTGTTGAATTTATTGAGTTGGAAAAGGGCATCGTAGATTTGATAAACCACCGCCGGGATAACCGAATAACGTACTCGACACCTTCGGCTTTGAGACTCTGCATATGATTTGCGCGCCTGATAACTGCATAACGACCTCTTTTATACGGTTTCAAACCAATAGGGACAAACCGCTTTGCCACAGCACTCGTAATGCAATTTCCACAGCAGCGATTAGGGTACGCGCATTTGGGGGAACACGGCAACAGACGGATTATCCAATCAATTGGAAAGGAACACAGAGTTTGTGAAAAAGAGTAGAAACGATAACCGCAACCGACAGTTCAATCAAGAAAAATCTTTCATCTTTTAAATATTTTGAAGCAGAGAAATATTGATTGATTTGATTTAAAGAAATAAAATCAGGAGTACCTTTTGAAGATGGAATTTGTGACAGTTTGTGTAGGAGGGGCAGATGTGAAAAACCTTCTTCGATATCAAGAATTGTAAATTTACAGGGTTTCATCCCAATAAAGACTCGGGATATTGATTGAACCTTGATTTTATTTTGATATATCAAAAATATCCCAACCATACTTCTGAAAATGGCTCATTCACCGAGCTGATTGGACGGCATTTGACAGAACCAGAGGCTAACAAACGACTTAATATATTGATGTATAGTGATTAACAAAATCAGGACAAGGCGACGAAGCTGCAGACAGTACAAATAGTACGGAACCGATTACTTGGTGCTTCAGCACCTTAGAGAATCGTCTCTTTGAGCTAAGCGAGGCAACGCGCTTACGTTTAAATTTAATCCACTATATTTAGTTTTATCTATTTTCAATTAACAGCAATAGACAAAAAATAACCGCTCTAAAAGCGGTTGTGGTGGCCAGGGTCGGACTCGAACCGACACACCTTGCGGCGGGGATTGTAGTCCCTCGCTTACCAATTTCCGCCACCTGGGCTGGTAAGACTCGCTATATAATGGCTTTGAAATTCGTAAACCTTTTTTTGAAATTTATTTATCTGTTTTATTTATTTTATTTTATTTTGAATTTTAAATAGAAATTTTATTTTAAATCTTACTGTTCTTCCGCTCCAAAGATTCTGTATGATTCCGGCAATTCCTGCCGTGCAGACAACGTAAAAAATACTACATTAATCTGCCAAACCGGTTAAGATGGAATATTCAAAATTCGTACGAATCAGGTTTTGCTATTTATTCTTGGGAGATTGTCATGTTTTCCGTACCGGTTCCTTTTTCGGGGCGTTTTCTGACTTGGCGCGCTTCCCGCTGCAAACTCAAGACAACAGTGCGCGCAAGTTCGTTCTTCAAGTGATCCGCGTGGCTGCGGAAAATGCGGCAAGCGCAACGCGCGGTACGGATATGCGTAAGGAAGACATCGCGCGGATTTACGCTGACCGACGGCAAGGCAAGCCTTTCAACCTGAGCGATTGAAAGGCAAGGTGCTGATTCTGTCTTTCCGGCTTACGCACTGTCCCGATGTCTGCCGACAGAGCTTTTGACGTACAGCGACAAGTTGAAGCAGTTGGCGGGCAGGCTAAGGACGTGAAAGTGGTGTTCGTCAGCATCGATCGGAACCGGACACCGCTGAATCATCGGCAAGTATGCCAAACAGTTCAATCCGGACTTTATCGGCTGTACGGCAACGGGGCGGCAAAACCTGCCGGTCATCAAGCAGCAATACCGCGTGGTTCTGCCAAAGTCAATCAAAAAGACGACAGCGAAACCTATTGTTGTCGACCACTCTCCGTGCGTATCTCATCGACAAAACGGTGAGGTTGCCATTTTCTCGCCTTACGGAAGCGAGCGGAAACGATTGCTGCCGATGTAAGGACCTGCTCTGATAAAACCGTATGCCGCTGCACCGTCCGGCGCTATTTCAGACGGCATTTGTTTCAACCGACAAAGGACATCCACACCATGCAGGATAATGCTTTGACCATCGCCTTATCCAAGGGCGCATTTTGGAGAGCGCTGCCGCTGCGCGTGCTGCGTAAAGGGTTTGATTACGAAGCAGCTTCGCAACCCGGATGCCGTCTGAAGATTGCCACAAAGTATCCTGAAATCGCGGCATCTATTTGCGCGCAAGGGTGTCCATGTGGACATTTATCAAACCTGTACGGCTCGATGGAACCTTGGCCCGCTGGTCCGCTTGACCGATGCGATTG

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CATACTATCCATAACATTATCATTTTATAGTATATAACAAAATGATTTTGCCCAAAAGTTT
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CTCGACAGTACGCTATTACGCTTTCTTTAAATGATGGCTGCTTCTAAGCCAAACATCCTG
GCTGCTGGGCTTCCACTTCTGTTTACCACTTAATCTATCATTTGGGACCTTAGCTGGC
GGTCTGGGTTGTTTCCCTCTTGACAACGGAGCTTAGCACCCGCTGTCTGCTCCCGAGGA

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ACCACCTGATGGTATTCTTAGTGGCCATGGGTTGGTAAAGTTGCAATAACCCCTAGCCA
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CTGCCGACCGCTGCTGCTGATGACATGAACGCTGCGCGCAAGATGCCATCGCATCAC
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ATATAAGAAATGCTGCACACAGGCATCCCCATGTGCAGCAGTTTTGATTTCAAAAGCGC
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CAAGTTCTGCCAACAGGTCTTTAAGCAGCAGCATTTTCTCGCGGCCAGCACTTCCTCGA
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CAGTCCGATGGAAGCAGACAGCAACTGGACACTTTCCAGTCCAGCCACACCGGCGCAC
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CATTGCCGTCGCCAAACTCGCCGGCCTGCCTGTACGCGCATTGAAATCCGCCCAAAGCA
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AAAACATTTTGAAGGTATATTGGCAGCAGCCTTGGAAAACTCGATCCCGACAGCCTGAC
CCCGCGCGAAGCATTGTCAGAACTGTACCGTCTGAAAGATTTGTGCAAATCCGTATCTTA
ATTTCCGTGTGTCGGAACAGCATCAAACCATATGGAAAAATCTGTGGATAAACATTATCTG
ACAGGAAATTTCCAAACATAAAAAATGCCGTCCGAACAGCTCAGACGGCATCCGTCCATT
CGGCT

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NMB Open Reading Frames

NMB0001 acetyltransferase, putative 491 3
NMB0002 hypothetical protein 890 498
NMB0003 glutamyl-tRNA synthetase 2305 914
NMB0004 EpiH/GdmH-related protein 3154 2513
NMB0005 arsenate reductase 3504 3154
NMB0006 thioredoxin-related protein 3628 4304
NMB0007 cell division ATP-binding protein FtsE 4304 4951
NMB0008 cell division protein FtsX, putative 4951 5865
NMB0009 BolA/YrbA family protein 5959 6204
NMB0010 phosphoglycerate kinase 7485 6277
NMB0011 UDP-N-acetylglucosamine 1-carboxyvinyltransferase 8819 7569
NMB0012 conserved hypothetical protein 10310 9342
NMB0013 conserved hypothetical protein 10792 10346
NMB0014 3-deoxy-D-manno-octulosonic-acid transferase 12104 10836
NMB0015 6-phosphogluconate dehydrogenase, decarboxylating 13615 12170
NMB0016 hypothetical protein 13911 14144
NMB0017 UDP-3-O-3-hydroxymyristoyl N-acetylglucosamine deacetylase 16137 15217
NMB0018 pilin PilE 17734 17225
NMB0019 pilS cassette 18932 18513
NMB0020 pilS cassette 19646 19263
NMB0021 pilS cassette 20297 19914
NMB0022 pilS cassette 21157 20894
NMB0023 pilS cassette 21882 21466
NMB0024 pilS cassette 22474 22061
NMB0025 large pilS cassette 23489 22821
NMB0026 pilS cassette 23868 23594
NMB0027 FKBP-type peptidyl-prolyl cis-trans isomerase 24226 23900
NMB0028 hypothetical protein 24522 24307
NMB0029 glycerate dehydrogenase 24644 25594
NMB0030 methionyl-tRNA synthetase 27729 25675
NMB0031 glucosamine--fructose-6-phosphate aminotransferase (isomerizing) 29683 27848
NMB0032 hypothetical protein 29959 30483
NMB0033 membrane-bound lytic murein transglycosylase A, putative 32229 30907
NMB0034 conserved hypothetical protein 32440 33276
NMB0035 conserved hypothetical protein 33276 34439
NMB0036 conserved hypothetical protein 34706 35968
NMB0037 phnA protein 36372 36046
NMB0038 UDP-N-acetylglucosamine pyrophosphorylase 37817 36450
NMB0039 hypothetical protein 38144 37875
NMB0040 hydrolase, putative 38850 38140
NMB0041 ABC transporter, periplasmic solute-binding protein 38909 39907
NMB0042 conserved hypothetical protein 40004 40849
NMB0043 conserved hypothetical protein 40878 41360
NMB0044 peptide methionine sulfoxide reductase 43033 41468
NMB0045 signal recognition particle protein 43179 44441
NMB0046 hypothetical protein 44451 44672
NMB0047 conserved hypothetical protein 45072 45353
NMB0048 conserved hypothetical protein FRAMESHIFT 47969 48109
NMB0049 pilC2 protein FRAMESHIFT 48116 51279
NMB0050 conserved hypothetical protein 55173 53026
NMB0051 twitching motility protein 56685 55462
NMB0052 twitching motility protein PilT 57891 56851
NMB0053 conserved hypothetical protein 58011 58694
NMB0054 hypothetical protein 58697 59101
NMB0055 pyrroline-5-carboxylate reductase 59153 59941

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NMB0056 DnaK suppressor protein 60091 60504
NMB0057 hypothetical protein 66347 66700
NMB0058 hypothetical protein 66731 66885
NMB0059 dnaJ protein 66972 68090
NMB0060 conserved hypothetical protein 68289 70304
NMB0061 dTDP-6-deoxy-L-lyxo-4-hexulose reductase FRAMESHIFT 70923 69924
NMB0062 glucose-1-phosphate thymidyltransferase 71828 70965
NMB0063 dTDP-D-glucose 4,6-dehydratase 72958 71894
NMB0064 UDP-glucose 4-epimerase 74093 73077
NMB0065 hypothetical protein 74476 75399
NMB0066 rRNA adenine N-6-methyltransferase 75687 76418
NMB0067 polysialic acid capsule biosynthesis protein SiaD, truncation
77283 76609
NMB0068 polysialic acid capsule biosynthesis protein SiaC 78416 77370
NMB0069 polysialic acid capsule biosynthesis protein SiaB 79103 78420
NMB0070 polysialic acid capsule biosynthesis protein synX 80240 79110
NMB0071 capsule polysaccharide export outer membrane protein CtrA 80375
81547
NMB0072 capsule polysaccharide export inner-membrane protein CtrB 81565
82725
NMB0073 capsule polysaccharide export inner-membrane protein CtrC 82728
83522
NMB0074 capsule polysaccharide export ATP-binding protein CtrD 83522 84169
NMB0075 transcriptional accessory protein Tex, putative 84236 86506
NMB0076 methyltransferase HphIm(C), FRAMESHIFT 86540 87539
NMB0077 site-specific DNA methylase, truncation 87529 87876
NMB0078 UDP-glucose 4-epimerase, truncation 87922 88575
NMB0079 dTDP-D-glucose 4,6-dehydratase 88694 89758
NMB0080 glucose-1-phosphate thymidyltransferase 89824 90687
NMB0081 dTDP-4-keto-6-deoxy-D-glucose-3,6-epimerase 90729 91280
NMB0082 capsule polysaccharide modification protein LipA 91308 93419
NMB0083 capsule polysaccharide modification protein LipB 93559 94815
NMB0084 conserved hypothetical protein FRAMESHIFT 95185 96587
NMB0085 sodium/glutamate symporter 96808 98019
NMB0086 hypothetical protein 98121 99134
NMB0087 hypothetical protein 99148 99342
NMB0088 outer membrane protein Pl, putative 101170 99773
NMB0089 pyruvate kinase II 102957 101488
NMB0090 IS1016 family transposase, putative FRAMESHIFT 103217 103857
NMB0091 hypothetical protein 104399 104632
NMB0092 hypothetical protein 104629 104853
NMB0093 hypothetical protein 104856 104939
NMB0094 hypothetical protein 105228 105413
NMB0095 hypothetical protein 105423 105572
NMB0096 hypothetical protein 105676 105843
NMB0097 secretion protein, putative POINT MUTATION 105860 107344
NMB0098 ABC transporter, ATP-binding protein FRAMESHIFT 107313 109396
NMB0099 hypothetical protein 109624 109484
NMB0100 hypothetical protein 109770 109627
NMB0101 IS1016 family transposase, putative FRAMESHIFT 109850 110489
NMB0102 hypothetical protein 110608 111123
NMB0103 bacteriocin resistance protein, putative 111896 111405
NMB0104 hypothetical protein 113073 112402
NMB0105 PhnO-related protein 114197 113358
NMB0106 aspartate carbamoyltransferase, catalytic subunit 114436 115353
NMB0107 aspartate carbamoyltransferase, regulatory subunit 115366 115821
NMB0108 hypothetical protein 115889 116551
NMB0109 conserved hypothetical protein 117948 116620
NMB0110 polypeptide deformylase 118018 118518
NMB0111 methionyl-tRNA formyltransferase 118608 119531
NMB0112 16S RNA methyltransferase 119613 120869
NMB0113 hypothetical protein 120892 121431
NMB0114 nitrogen regulation protein NtrY, putative 121434 123551
NMB0115 nitrogen assimilation regulatory protein NtrX 123547 124821

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NMB0116 DNA processing chain A 124915 126105
NMB0117 smg protein, putative 126134 126592
NMB0118 DNA topoisomerase I 126667 128970
NMB0119 hypothetical protein 129741 129049
NMB0120 hypothetical protein 130312 129764
NMB0121 conserved hypothetical protein 130431 130805
NMB0122 conserved hypothetical protein 130897 131463
NMB0123 ferredoxin, 4Fe-4S bacterial type 131589 131837
NMB0124 translation elongation factor Tu 132257 133438
NMB0125 preprotein translocase subunit SecE 133638 133913
NMB0126 transcription antitermination protein NusG 133918 134451
NMB0127 50S ribosomal protein L11 134555 134986
NMB0128 50S ribosomal protein L1 134989 135681
NMB0129 hypothetical protein 135753 135893
NMB0130 50S ribosomal protein L10 135914 136411
NMB0131 50S ribosomal protein L7/L12 136472 136840
NMB0132 DNA-directed RNA polymerase, beta subunit FRAMESHIFT 137027 141208
NMB0133 DNA-directed RNA polymerase, beta' subunit 141368 145540
NMB0134 hypothetical protein 145835 146089
NMB0135 conserved hypothetical protein 146089 146235
NMB0136 30S ribosomal protein S12 146417 146785
NMB0137 30S ribosomal protein S7 146906 147373
NMB0138 elongation factor G (EF-G) 147395 149497
NMB0139 translation elongation factor Tu 149586 150767
NMB0140 30S ribosomal protein S10 150788 151096
NMB0141 transposase, truncation 151241 151603
NMB0142 50S ribosomal protein L3 151777 152418
NMB0143 50S ribosomal protein L4 152421 153038
NMB0144 50S ribosomal protein L23 153038 153349
NMB0145 50S ribosomal protein L2 153358 154188
NMB0146 30S ribosomal protein S19 154198 154473
NMB0147 50S ribosomal protein L22 154485 154811
NMB0148 30S ribosomal protein S3 154824 155513
NMB0149 50S ribosomal protein L16 155500 155913
NMB0150 50S ribosomal protein L29 155916 156104
NMB0151 30S ribosomal protein S17 156107 156367
NMB0152 50S ribosomal protein L14 156592 156957
NMB0153 50S ribosomal protein L24 156972 157292
NMB0154 50S ribosomal protein L5 157305 157841
NMB0155 30S ribosomal protein S14 157847 158149
NMB0156 30S ribosomal protein S8 158168 158557
NMB0157 50S ribosomal protein L6 158574 159104
NMB0158 50S ribosomal protein L18 159121 159471
NMB0159 30S ribosomal protein S5 159493 160008
NMB0160 50S ribosomal protein L30 160004 160186
NMB0161 50S ribosomal protein L15 160191 160622
NMB0162 preprotein translocase SecY subunit 160637 161944
NMB0163 translation initiation factor IF-1 161952 162167
NMB0164 50S ribosomal protein L36 162191 162301
NMB0165 30S ribosomal protein S13 162370 162729
NMB0166 30S ribosomal protein S11 162752 163144
NMB0167 30S ribosomal protein S4 163167 163784
NMB0168 DNA-directed RNA polymerase, alpha subunit 163813 164796
NMB0169 50S ribosomal protein L17 164823 165188
NMB0170 septum site-determining protein MinC 165338 166048
NMB0171 septum site-determining protein MinD 166079 166891
NMB0172 cell division topological specificity factor 166898 167158
NMB0173 transcriptional regulator, LysR family 167165 168082
NMB0174 valyl-tRNA synthetase 171252 168418
NMB0175 conserved hypothetical protein 172158 171352
NMB0176 D-amino acid dehydrogenase, small subunit 173595 172342
NMB0177 sodium/alanine symporter, putative 175065 173677
NMB0178 acyl-(acyl-carrier-protein)--UDP-N-acetylglucosamine O-acyltransferase 176198 175425

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NMB0179 (3R)-hydroxymyristoyl-(acyl carrier protein) dehydratase 176734
176288
NMB0180 UDP-3-O-(3-hydroxymyristoyl)-glucosamine N-acyltransferase 177814
176771
NMB0181 outer membrane protein OmpH, putative 178347 177850
NMB0182 outer membrane protein Omp85 180806 178416
NMB0183 conserved hypothetical protein 182203 180866
NMB0184 1-deoxy-D-xylulose 5-phosphate reductoisomerase 183422 182241
NMB0185 phosphatidate cytidyltransferase 184275 183481
NMB0186 undecaprenyl pyrophosphate synthetase 185024 184281
NMB0187 ribosome recycling factor 185637 185083
NMB0188 conserved hypothetical protein 186944 185820
NMB0189 hypothetical protein 187355 187774
NMB0190 glucose inhibited division protein B 187935 188555
NMB0191 ParA family protein 188657 189427
NMB0192 ribonuclease HII 191274 190693
NMB0193 glucose inhibited division protein A 193238 191346
NMB0194 amino acid symporter, putative 194991 193567
NMB0195 pyridoxal phosphate biosynthetic protein PdxA 195133 196137
NMB0196 ribonuclease E 200197 197441
NMB0197 hypothetical protein 200321 200605
NMB0198 ribosomal large subunit pseudouridine synthase C 200690 201679
NMB0199 lipid-A-disaccharide synthase 201730 202899
NMB0200 hypothetical protein 203501 203115
NMB0201 hypothetical protein 203724 204131
NMB0202 hypothetical protein 204152 204322
NMB0203 dihydrodipicolinate reductase 205207 204401
NMB0204 lipoprotein, putative 205594 205220
NMB0205 ferric uptake regulation protein 205813 206244
NMB0206 leucyl/phenylalanyl-tRNA--protein transferase 206317 207039
NMB0207 glyceraldehyde 3-phosphate dehydrogenase 208326 207298
NMB0208 ferredoxin, 4Fe-4S bacterial type 209364 208528
NMB0209 glutathione-regulated potassium-efflux system protein 209513
211486
NMB0210 site-specific DNA methylase, truncation 212082 212401
NMB0211 L-serine dehydratase 214093 212711
NMB0212 DNA gyrase subunit B 216580 214193
NMB0213 hypothetical protein 216736 217719
NMB0214 oligopeptidase A 217810 219843
NMB0215 conserved hypothetical protein 221035 220472
NMB0216 catalase 222945 221434
NMB0217 RNA polymerase sigma-54 factor RpoN, putative 223293 224141
NMB0218 glycosyltransferase 226194 225067
NMB0219 3-oxoacyl-(acyl-carrier-protein) synthase II 227746 226502
NMB0220 acyl carrier protein 228138 227905
NMB0221 dihydroorotate dehydrogenase 228370 229374
NMB0222 hypothetical protein 229540 230010
NMB0223 hypothetical protein 230140 230355
NMB0224 glutamate-ammonia-ligase adenylyltransferase 230556 233243
NMB0225 transposase, IS30 family FRAMESHIFT 234513 233551
NMB0226 conserved hypothetical protein 235470 234781
NMB0227 conserved hypothetical protein 236771 235581
NMB0228 conserved hypothetical protein 237637 236903
NMB0229 conserved hypothetical protein FRAMESHIFT 238552 237662
NMB0230 conserved hypothetical protein 239196 238552
NMB0231 hypothetical protein 239356 239255 N
NMB0232 DNA helicase II 239380 241584
NMB0233 hypothetical protein 241663 241761
NMB0234 hypothetical protein 242111 242647
NMB0235 hypothetical protein 243052 242894
NMB0236 hypothetical protein 243168 243063
NMB0237 hypothetical protein 243535 243179
NMB0238 IS1016 family transposase, degenerate 243588 243849
NMB0239 hypothetical protein 244051 244668

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NMB0240 hypothetical protein 244694 246142
NMB0241 NADH dehydrogenase I, A subunit 246607 246960
NMB0242 NADH dehydrogenase I, B subunit 246954 247433
NMB0243 NADH dehydrogenase I, C subunit 247449 248039
NMB0244 NADH dehydrogenase I, D subunit 248032 249285
NMB0245 NADH dehydrogenase I, E subunit 249288 249758
NMB0246 NADH dehydrogenase I, F subunit 250151 251449
NMB0247 hypothetical protein 251452 251886
NMB0248 conserved hypothetical protein 252175 252411
NMB0249 NADH dehydrogenase I, G subunit 252726 254984
NMB0250 NADH dehydrogenase I, H subunit 254990 256063
NMB0251 NADH dehydrogenase I, I subunit 256147 256623
NMB0252 hypothetical protein 256657 257361
NMB0253 NADH dehydrogenase I, J subunit 257400 258068
NMB0254 NADH dehydrogenase I, K subunit 258068 258370
NMB0255 cell filamentation protein Fic-related protein 258407 258979
NMB0256 hypothetical protein 259106 259444
NMB0257 NADH dehydrogenase I, L subunit 259496 261517
NMB0258 NADH dehydrogenase I, M subunit 261616 263109
NMB0259 NADH dehydrogenase I, N subunit 263122 264561
NMB0260 hypothetical protein 264612 264995
NMB0261 geranyltranstransferase 265863 265087
NMB0262 exodeoxyribonuclease, small subunit 266188 265967
NMB0263 conserved hypothetical protein 267358 266438
NMB0264 ABC transporter, ATP-binding protein 269219 267366
NMB0265 Holliday junction DNA helicase RuvA 269966 269385
NMB0266 conserved hypothetical protein 270374 270051
NMB0267 conserved hypothetical protein 271155 270439
NMB0268 RNA methyltransferase, TrmH family 271749 271288
NMB0269 competence protein 272539 271817
NMB0270 bioH protein, putative 272538 273284
NMB0271 hypothetical protein 273284 274069
NMB0272 hypothetical protein 274527 274820
NMB0273 hypothetical protein 274861 275283
NMB0274 ATP-dependent DNA helicase RecQ 277728 275431
NMB0275 indole-3-glycerol phosphate synthase 278575 277796
NMB0276 conserved hypothetical protein 279582 278629
NMB0277 virulence factor MviN 281255 279717
NMB0278 thiol:disulfide interchange protein DsbA 281470 282165
NMB0279 conserved hypothetical protein 283229 282228
NMB0280 organic solvent tolerance protein, putative 283431 285704
NMB0281 peptidyl-prolyl cis-trans isomerase 285809 286852
NMB0282 ribonuclease II-related protein 290243 288366
NMB0283 conserved hypothetical protein 290552 291181
NMB0284 adenylosuccinate lyase 291256 292623
NMB0285 O-antigen acetylase FRAMESHIFT 292707 294573
NMB0286 conserved hypothetical protein 295481 294870
NMB0287 probable ATP-dependent helicase DinG 297668 295521
NMB0288 hypothetical protein 297740 297967
NMB0289 deoxyribodipyrimidine photolyase, FRAMESHIFT 299363 298066
NMB0290 transcriptional regulator, putative 300264 299356
NMB0291 conserved hypothetical protein 300372 300767
NMB0292 conserved hypothetical protein 300819 301421
NMB0293 TonB-dependent receptor, putative 301610 303718
NMB0294 thiol:disulfide interchange protein DsbA 303836 304528
NMB0295 signal recognition particle protein 306232 304865
NMB0296 CcsA-related protein 306452 307255
NMB0297 hypothetical protein 307272 307367
NMB0298 hypothetical protein 307401 307583
NMB0299 comEA-related protein 313097 313540
NMB0300 hypothetical protein 313603 313904
NMB0301 Hypothetical protein 313958 314161
NMB0302 IS1016C2 transposase, degenerate 314284 314933
NMB0303 transposase, degenerate 315024 315307

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NMB0304 class 5 outer membrane protein, degenerate 315549 315295
NMB0305 hypothetical protein 315891 315736
NMB0306 hypothetical protein 316061 316252
NMB0307 phospho-2-dehydro-3-deoxyheptonate aldolase, phe-sensitive 316403
317455
NMB0308 dihydrofolate reductase 317526 318011
NMB0309 conserved hypothetical protein 318840 318367
NMB0310 conserved hypothetical protein 319280 318855
NMB0311 hypothetical protein 319392 319634
NMB0312 virulence-associated protein VapA FRAMESHIFT 321089 323177
NMB0313 conserved hypothetical protein 323422 324885
NMB0314 hypothetical protein 326057 325092
NMB0315 conserved hypothetical protein 326135 327424
NMB0316 conserved hypothetical protein 328616 327933
NMB0317 conserved hypothetical protein 329164 328694
NMB0318 fatty acid efflux system protein 329606 330757
NMB0319 fatty acid efflux system protein 330784 332307
NMB0320 hypothetical protein 332373 332519
NMB0321 50S ribosomal protein L28 332560 332790
NMB0322 50S ribosomal protein L33 332825 332977
NMB0323 UbiH family protein 334353 333172
NMB0324 50S ribosomal protein L27 334964 334695
NMB0325 50S ribosomal protein L21 335297 334992
NMB0326 octaprenyl-diphosphate synthase 335521 336492
NMB0327 conserved hypothetical protein FRAMESHIFT 336500 336944
NMB0328 hypothetical protein 336993 337165
NMB0329 type IV pilus assembly protein 337388 339061
NMB0330 conserved hypothetical protein 339358 339152
NMB0331 kinase, putative 339983 339354
NMB0332 type IV prepilin peptidase 340845 339988
NMB0333 pilus assembly protein PilG 342151 340922
NMB0334 glucose-6-phosphate isomerase 342508 344148
NMB0335 2,3,4,5-tetrahydropyridine-2-carboxylate N-succinyltransferase
344361 345179
NMB0336 enoyl-(acyl-carrier-protein) reductase 345337 346119
NMB0337 branched-chain amino acid aminotransferase, putative 347364 346369
NMB0338 hypothetical protein 347506 347985
NMB0339 conserved hypothetical protein 347999 349165
NMB0340 lactoylglutathione lyase FRAMESHIFT 349193 349605
NMB0341 tspA protein 352407 349783
NMB0342 intracellular septation protein A 352613 353140
NMB0343 conserved hypothetical protein 353158 353433
NMB0344 BolA/YrbA family protein 353436 353711
NMB0345 cell-binding factor, putative 353763 354626
NMB0346 hypothetical protein 354700 355455
NMB0347 conserved hypothetical protein 355531 356019
NMB0348 conserved hypothetical protein 356053 357060
NMB0349 glutamyl-tRNA synthetase-related protein 358020 357136
NMB0350 hypothetical protein 358760 358311
NMB0351 transaldolase 359966 358914
NMB0352 sugar isomerase, KpsF/GutQ family 360063 361034
NMB0353 conserved hypothetical protein 361255 361788
NMB0354 hypothetical protein 361788 362366
NMB0355 conserved hypothetical protein 362350 362877
NMB0356 ABC transporter, ATP-binding protein 362924 363685
NMB0357 monofunctional biosynthetic peptidoglycan transglycosylase 364858
364160
NMB0358 shikimate 5-dehydrogenase 365670 364864
NMB0359 glutamate--ammonia ligase 365970 367385
NMB0360 AmpG-related protein 367544 368824
NMB0361 conserved hypothetical protein 368824 369096
NMB0362 hypothetical protein 369205 369282
NMB0363 hypothetical protein 369610 369744
NMB0364 FrpC operon protein 370088 370858

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NMB0365 iron-regulated protein FrpC, truncation 370878 371150
NMB0366 hypothetical protein 372373 371243
NMB0367 hypothetical protein 372823 372440
NMB0368 hypothetical protein 373350 372895
NMB0369 hypothetical protein 373720 373334
NMB0370 hypothetical protein 374229 373855
NMB0371 hypothetical protein 374658 374254
NMB0372 hypothetical protein 375341 374667
NMB0373 hypothetical protein 375915 375559
NMB0374 MafB-related protein 377321 375921
NMB0375 mafA protein 378266 377328
NMB0376 hypothetical protein 378379 378266
NMB0377 conserved hypothetical protein 379516 378389
NMB0378 phosphate permease, putative 379807 381378
NMB0379 oxygen-independent coproporphyrinogen III oxidase 383155 381737
NMB0380 transcriptional regulator, Crp/Fnr family 383360 384091
NMB0381 cys regulon transcriptional activator 385157 384210
NMB0382 outer membrane protein class 4 385521 386246
NMB0383 hypothetical protein 386270 386494
NMB0384 hypothetical protein 386773 387066
NMB0385 thiamin-monophosphate kinase 387100 388053
NMB0386 phosphatidylglycerophosphatase A 388049 388531
NMB0387 ABC transporter, ATP-binding protein 390270 388597
NMB0388 sugar transporter, putative 390657 392009
NMB0389 aldose 1-epimerase 392016 393023
NMB0390 maltose phosphorylase 393260 395515
NMB0391 beta-phosphoglucomutase 395531 396193
NMB0392 l-aspartate oxidase 397882 396377
NMB0393 multidrug resistance protein 398266 397934
NMB0394 quinolinate synthetase A 399530 398421
NMB0395 conserved hypothetical protein 399732 400667
NMB0396 nicotinate-nucleotide pyrophosphorylase 400888 401766
NMB0397 hypothetical protein 401797 402081
NMB0398 transcriptional regulator, ArsR family 402176 402454
NMB0399 exodeoxyribonuclease III 402517 403284
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NMB0401 proline dehydrogenase 409441 405839
NMB0402 sodium/proline symporter 411216 409693
NMB0403 hypothetical protein 411644 411555
NMB0404 conserved hypothetical protein 411699 412016
NMB0405 competence protein ComM 412033 413526
NMB0406 conserved hypothetical protein 413629 414495
NMB0407 thiol:disulfide interchange protein DsbA 414501 415142
NMB0408 bacitracin resistance protein 415178 415996
NMB0409 conserved hypothetical protein 417783 416575
NMB0410 conserved hypothetical protein 418062 418514
NMB0411 conserved hypothetical protein 418514 419497
NMB0412 cell division protein FtsL-related protein 419491 419757
NMB0413 penicillin-binding protein 2 419821 421563
NMB0414 UDP-N-acetylmuramoylalanyl-D-glutamate--2,6-diaminopimelate ligase
421591 423066
NMB0415 conserved hypothetical protein FRAMESHIFT 423092 424736
NMB0416 UDP-N-acetylmuramoylalanyl-D-glutamyl-2,6-diaminopimelate--D-
alanyl-D- alanyl ligase 424864 426228
NMB0417 hypothetical protein 426234 426407
NMB0418 phospho-N-acetylmuramoyl-pentapeptide-transferase 426657 427736
NMB0419 conserved hypothetical protein 427865 428458
NMB0420 UDP-N-acetylmuramoylalanine--D-glutamate ligase 428545 429879
NMB0421 cell division protein FtsW 430062 431330
NMB0422 UDP-N-acetylglucosamine--N-acetylmuramyl-(pentapeptide)
pyrophosphoryl-undecaprenol N-acetylglucosamine transferase
431337 432401
NMB0423 UDP-N-acetylmuramate--alanine ligase 432559 433965
NMB0424 D-alanine--D-alanine ligase 434081 434992

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NMB0425 cell division protein FtsQ 435006 435710
NMB0426 cell division protein FtsA 435799 437040
NMB0427 cell division protein FtsZ 437162 438337
NMB0428 conserved hypothetical protein 438479 439786
NMB0429 hypothetical protein 440162 440263
NMB0430 carboxyphosphoenolpyruvate phosphonmutase, putative 440412
441287
NMB0431 methylcitrate synthase/citrate synthase 2 441376 442527
NMB0432 conserved hypothetical protein 442683 443468
NMB0433 aconitate hydratase 1 443549 446152
NMB0434 conserved hypothetical protein 446958 448124
NMB0435 acetate kinase 448541 449737
NMB0436 conserved hypothetical protein 450078 450716
NMB0437 conserved hypothetical protein 451289 450849
NMB0438 hypothetical protein 451463 451828
NMB0439 conserved hypothetical protein 451876 453027
NMB0440 prephenate dehydrogenase, putative 453959 453090
NMB0441 nitrilase 454044 454853
NMB0442 opacity protein FRAMESHIFT 455681 454888
NMB0443 transposase, IS30 family 456456 457418
NMB0444 conserved hypothetical protein 457979 458830
NMB0445 bicyclomycin resistance protein, putative 459352 460581
NMB0446 chorismate mutase/prephenate dehydratase 460662 461747
NMB0447 DNA repair protein RecO 461787 462575
NMB0448 pyridoxal phosphate biosynthetic protein PdxJ 462602 463327
NMB0449 hypothetical protein 463482 463703
NMB0450 hypothetical protein 463968 464411
NMB0451 hypothetical protein 464424 465188
NMB0452 holo-(acyl-carrier protein) synthase 465391 465765
NMB0453 mutT protein 465850 466656
NMB0454 hypothetical protein 466652 467071
NMB0455 conserved hypothetical protein 467123 468262
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NMB0457 conserved hypothetical protein 470031 469573
NMB0458 glutamate racemase 470233 471042
NMB0459 conserved hypothetical protein 473202 472096
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NMB0461 transferrin-binding protein 1 477798 480542
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spermidine/putrescine-binding protein 483195 481819
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NMB0464 phospholipase A1, putative 483685 484830
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NMB0467 hypothetical protein 487694 487975
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NMB0469 agmatinase 490136 491056
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NMB0471 conserved hypothetical protein 494006 492933
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NMB0476 hypothetical protein 498457 499551
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NMB0478 hypothetical protein 500104 500745
NMB0479 conserved hypothetical protein 500771 501127
NMB0480 TspB-related protein 502193 501801
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NMB0482 hypothetical protein 502900 502625
NMB0483 Hypothetical protein 503191 502910
NMB0484 hypothetical protein 503396 503202
NMB0485 hypothetical protein 503691 503404
NMB0486 conserved hypothetical protein FRAMESHIFT 505078 503739

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NMB0487 hypothetical protein 505244 505152
NMB0488 hypothetical protein 505800 505309
NMB0489 hypothetical protein 506682 505804
NMB0490 PspA-related protein 507809 506910
NMB0491 hypothetical protein 508744 508304
NMB0492 hypothetical protein 509383 509063
NMB0493 hemagglutinin/hemolysin-related protein 517494 509386
NMB0494 DNA helicase, truncation 518107 517625
NMB0495 replication protein 519187 518207
NMB0496 hemolysin activator-related protein 519134 520810
NMB0497 hemagglutinin/hemolysin-related protein 520922 526826
NMB0498 hypothetical protein 526836 527342
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NMB0500 hypothetical protein 529102 529476
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NMB0503 hypothetical protein 532134 532562
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NMB0507 hypothetical protein 535208 535693
NMB0508 hypothetical protein 535883 536152
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NMB0510 hypothetical protein 537136 537396
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NMB0519 hypothetical protein 542725 542925
NMB0520 hypothetical protein 542931 543107
NMB0521 hypothetical protein 543492 543947
NMB0522 transposase, truncated 543958 544080
NMB0523 ABC transporter, ATP-binding protein, truncation 544162 544441
NMB0524 ribonuclease BN, putative 545691 544474
NMB0525 aluminum resistance protein, putative 546236 546892
NMB0526 hypothetical protein 546923 547438
NMB0527 6-pyruvoyl tetrahydrobiopterin synthase, putative 547448 547867
NMB0528 conserved hypothetical protein 548139 548507
NMB0529 conserved hypothetical protein 548507 549142
NMB0530 glycosyl hydrolase, family 3 550869 549787
NMB0531 conserved hypothetical protein 552446 550929
NMB0532 protease DO 554147 552651
NMB0533 endonuclease III 554914 554288
NMB0534 conserved hypothetical protein 555373 554963
NMB0535 glucose/galactose transporter 555906 557183
NMB0536 Na⁺/H⁺ antiporter 557477 558853
NMB0537 conserved hypothetical protein 559809 558988
NMB0538 conserved hypothetical protein 560326 559820
NMB0539 porphobilinogen deaminase 560445 561377
NMB0540 aspartate aminotransferase 562977 561787
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NMB0543 L-lactate permease, putative 565630 564047
NMB0544 conserved hypothetical protein 566621 565902
NMB0545 conserved hypothetical protein 566870 570352
NMB0546 alcohol dehydrogenase, propanol-preferring 571566 570523
NMB0547 type IV pilin protein 572238 571852
NMB0548 AcrA/AcrE family protein 572464 573639
NMB0549 ABC transporter, ATP-binding protein 573708 575639
NMB0550 thiol:disulfide interchange protein DsbC 576837 576058
NMB0551 primosomal protein n' 576975 579161

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NMB0552 hypothetical protein 580284 579214
NMB0553 transposase, putative, POINT MUTATION 581288 580335
NMB0554 dnaK protein 584451 582526
NMB0555 hypothetical protein 584931 584662
NMB0556 repressor protein, putative 585119 585802
NMB0557 conserved hypothetical protein 585937 586272
NMB0558 hypothetical protein 586435 586896
NMB0559 ubiquinone biosynthesis protein AarF 586934 588442
NMB0560 serine acetyltransferase 589620 588805
NMB0561 grpE protein 589804 590379
NMB0562 conserved hypothetical protein 590874 590662
NMB0563 thiamine biosynthesis lipoprotein ApbE 591955 590903
NMB0564 Na(+)-translocating NADH-quinone reductase, subunit F 593325 592111
NMB0565 Na(+)-translocating NADH-quinone reductase, subunit E 593932 593342
NMB0566 Na(+)-translocating NADH-quinone reductase, subunit D 594562 593939
NMB0567 Na(+)-translocating NADH-quinone reductase, subunit C 595338 594565
NMB0568 Na(+)-translocating NADH-quinone reductase, subunit B 596563 595334
NMB0569 Na(+)-translocating NADH-quinone reductase, subunit A 597909 596569
NMB0570 hypothetical protein 599680 598262
NMB0571 conserved hypothetical protein 600400 600044
NMB0572 hypothetical protein 601002 600400
NMB0573 transcriptional regulator, AsnC family 601612 601052
NMB0574 glycine cleavage system T protein 602042 603139
NMB0575 glycine cleavage system H protein 603304 603687
NMB0576 glutamyl-tRNA reductase 603842 605086
NMB0577 NosR-related protein 605365 605934
NMB0578 copper ABC transporter, periplasmic copper-binding protein 605991 607022
NMB0579 copper ABC transporter, ATP-binding protein 607083 607700
NMB0580 protein disulfide isomerase NosL, putative 607842 608333
NMB0581 electron transfer flavoprotein-ubiquinone oxidoreductase 610085 608427
NMB0582 bacteriocin resistance protein, putative 610757 610218
NMB0583 IS1016C2 transposase 612651 611986
NMB0584 FrpC operon protein 613242 614054
NMB0585 iron-regulated protein FrpA, putative 614074 617979
NMB0586 adhesin, putative 619176 618265
NMB0587 membrane protein 620128 619256
NMB0588 ABC transporter, ATP-binding protein 620907 620155
NMB0589 50s ribosomal protein L19 621563 621201
NMB0590 tRNA (guanine-N1)-methyltransferase FRAMESHIFT 622329 621582
NMB0591 16S rRNA processing protein RimM 622838 622332
NMB0592 30S ribosomal protein S16 623099 622857
NMB0593 conserved hypothetical protein 625570 623147
NMB0594 sensor histidine kinase 627094 625691
NMB0595 DNA-binding response regulator 627785 627111
NMB0596 hypothetical protein 629789 627978
NMB0597 hypothetical protein 630132 629782
NMB0598 Maf/YceF/YhdE family protein 630749 630144
NMB0599 conserved hypothetical protein 631572 630805
NMB0600 hypothetical protein 632272 631589
NMB0601 conserved hypothetical protein 632479 632279
NMB0602 hitA protein 632849 632529
NMB0603 phosphoribosyl-ATP cyclohydrolase 633244 632924
NMB0604 alcohol dehydrogenase, zinc-containing 634449 633388
NMB0605 histone deacetylase family protein 636107 635001
NMB0606 conserved hypothetical protein 636235 636498
NMB0607 protein-export membrane protein SecD 636710 638563

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NMB0608 protein-export membrane protein SecF 638570 639502
NMB0609 30s ribosomal protein S15 639728 639994
NMB0610 spermidine/putrescine ABC transporter, ATP-binding protein 640243
641499
NMB0611 spermidine/putrescine ABC transporter, permease protein 641518
642480
NMB0612 spermidine/putrescine ABC transporter, permease protein 642483
643367
NMB0613 hypothetical protein 643392 643496
NMB0614 oxidoreductase, putative 643496 644788
NMB0615 ammonium transporter AmtB, putative 646340 645039
NMB0616 IS1016 family transposase, degenerate 647272 646871
NMB0617 transcription termination factor Rho 648837 647581
NMB0618 phosphoenolpyruvate synthase 651441 649060
NMB0619 conserved hypothetical protein 651853 652671
NMB0620 phosphoglycolate phosphatase 653575 652916
NMB0621 conserved hypothetical protein 654440 653616
NMB0622 outer membrane lipoprotein carrier protein 654867 655487
NMB0623 spermidine/putrescine ABC transporter, periplasmic
spermidine/putrescine-binding protein 655763 656899
NMB0624 galactosyltransferase-related protein FRAMESHIFT 657035 658253
NMB0625 conserved hypothetical protein 658297 658824
NMB0626 peptide chain release factor 3 660797 659205
NMB0627 phosphoribosyl-AMP cyclohydrolase 661299 660907
NMB0628 hisF protein 662097 661333
NMB0629 phosphoribosylformimino-5-aminoimidazole carboxamide ribotide
isomerase 662847 662113
NMB0630 amidotransferase HisH 663518 662883
NMB0631 phosphate acetyltransferase Pta, putative 665151 663652
NMB0632 iron(III) ABC transporter, ATP-binding protein 666394 665339
NMB0633 iron(III) ABC transporter, permease protein 667932 666418
NMB0634 iron(III) ABC transporter, periplasmic binding protein 668995
668003
NMB0635 transposase, IS30 family 670247 669285
NMB0636 hypothetical protein 670794 670414
NMB0637 argininosuccinate lyase 672228 670855
NMB0638 UTP--glucose-1-phosphate uridylyltransferase 673116 672250
NMB0639 conserved hypothetical protein 673743 673147
NMB0640 hypothetical protein 673969 673739
NMB0641 inorganic pyrophosphatase 674610 674080
NMB0642 dATP pyrophosphohydrolase 675169 674714
NMB0643 MafB-related protein 675614 677437
NMB0644 hypothetical protein 677443 677904
NMB0645 ribonuclease FRAMESHIFT 677948 678275
NMB0646 ribonuclease inhibitor barstar 678290 678574
NMB0647 hypothetical protein 679091 680326
NMB0648 hypothetical protein 680357 680776
NMB0649 hypothetical protein 680970 681191
NMB0650 hypothetical protein 681167 681583
NMB0651 hypothetical protein 681687 682073
NMB0652 mafA protein 682199 683137
NMB0653 MafB-related protein 683144 684409
NMB0654 hypothetical protein 684415 684729
NMB0655 hypothetical protein 684867 685571
NMB0656 hypothetical protein 685600 685926
NMB0657 hypothetical protein 686024 686224
NMB0658 Hypothetical protein 686055 686312
NMB0659 hypothetical protein 686346 686744
NMB0660 hypothetical protein 686929 687315
NMB0661 bis(5'-nucleosyl)-tetrakisphosphatase, symmetrical/Trk system
potassium uptake protein TrkG FRAMESHIFT 689659 687362
NMB0662 ribonuclease, putative 690126 689740
NMB0663 outer membrane protein NsgA 690786 690265
NMB0664 hypothetical protein 691151 690960

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NMB0665 oxygen-independent coprophorphyrinogen III oxidase family protein
692546 691374
NMB0666 DNA ligase 695128 692606
NMB0667 hypothetical protein 696562 695279
NMB0668 ampD protein 697352 696783
NMB0669 conserved hypothetical protein 697436 698428
NMB0670 thymidylate kinase 698491 699108
NMB0671 malate oxidoreductase (NAD) 699333 700610
NMB0672 tetraacyldisaccharide 4'-kinase 701160 702191
NMB0673 hypothetical protein 702394 702978
NMB0674 conserved hypothetical protein 703050 703229
NMB0675 3-deoxy-D-manno-octulosonate cytidyltransferase 703229 703987
NMB0676 hypothetical protein 704013 704411
NMB0677 hypothetical protein 704610 704723
NMB0678 tryptophan synthase, alpha subunit 705306 706088
NMB0679 acetyl-CoA carboxylase, carboxyl transferase beta subunit 706129
706998
NMB0680 cryptic protein 707672 707064
NMB0681 conserved hypothetical protein 707781 708002
NMB0682 dihydroorotase 708368 709399
NMB0683 N utilization substance protein B 710195 709773
NMB0684 riboflavin synthase, beta subunit 710749 710276
NMB0685 hypothetical protein 711120 710800
NMB0686 ribonuclease III 711287 712003
NMB0687 GTP-binding protein Era 712003 712974
NMB0688 N-(5'-phosphoribosyl)anthranilate isomerase 715446 714823
NMB0689 transcription elongation factor GreB 715996 715508
NMB0690 amidophosphoribosyltransferase 717640 716099
NMB0691 colicin V production protein, putative 718450 717956
NMB0692 tpc protein 719441 718446
NMB0693 folylpolyglutamate synthase/dihydrofolate synthase 720728 719457
NMB0694 folI protein 721205 720762
NMB0695 hypothetical protein 721569 721213
NMB0696 amino acid ABC transporter, ATP-binding protein FRAMESHIFT 722369
721645
NMB0697 dimethyladenosine transferase 723321 722545
NMB0698 hypothetical protein 723518 724204
NMB0699 tryptophan synthase, beta subunit 724290 725489
NMB0700 IgA-specific serine endopeptidase 731118 725674
NMB0701 hypothetical protein 731531 731280
NMB0702 competence protein ComA 732529 734601
NMB0703 competence lipoprotein ComL 735635 734835
NMB0704 ribosomal large subunit pseudouridine synthase D 735634 736755
NMB0705 transporter 737858 736914
NMB0706 conserved hypothetical protein 738418 739194
NMB0707 rare lipoprotein B, putative 739249 739725
NMB0708 DNA polymerase III, delta subunit 739730 740725
NMB0709 Hypothetical protein 740849 741265
NMB0710 Hypothetical protein 741293 741856
NMB0711 conserved hypothetical protein FRAMESHIFT 742826 741946
NMB0712 RNA polymerase sigma-32 factor 744182 743313
NMB0713 apolipoprotein N-acyltransferase, putative 746012 744441
NMB0714 conserved hypothetical protein FRAMESHIFT 746771 746019
NMB0715 Hypothetical protein 746967 747284
NMB0716 Hypothetical protein 747440 747727
NMB0717 cytochrome, putative 748209 747796
NMB0718 ferrochelatase 749572 748493
NMB0719 queuine tRNA-ribosyltransferase 750697 749585
NMB0720 threonyl-tRNA synthetase 751005 752915
NMB0721 translation initiation factor 3 752990 753454
NMB0722 50S ribosomal protein L35 753604 753798
NMB0723 50S ribosomal protein L20 753814 754170
NMB0724 phenylalanyl-tRNA synthetase, alpha chain 754519 755508
NMB0725 modification methylase HgaI-1 755694 756749

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NMB0726 type II restriction enzyme HgaI 756755 758221
NMB0727 N-6 adenine-specific DNA methylase 758221 758868
NMB0728 phenylalanyl-tRNA synthetase, beta chain 758896 761256
NMB0729 integration host factor, alpha subunit 761333 761632
NMB0730 hypothetical protein 762257 762739
NMB0731 hypothetical protein 763002 763226
NMB0732 adenosylmethionine-8-amino-7-oxononanoate aminotransferase 763559
764857
NMB0733 dethiobiotin synthase 764857 765501
NMB0734 hypothetical protein 765519 765992
NMB0735 4-hydroxybenzoate octaprenyltransferase 766025 766912
NMB0736 PTS system, nitrogen regulatory IIA protein 767100 767546
NMB0737 HPr kinase/phosphatase, putative 767551 768510
NMB0738 conserved hypothetical protein 768494 769345
NMB0739 conserved hypothetical protein 769429 770943
NMB0740 DNA repair protein RecN 771255 772925
NMB0741 conserved hypothetical protein 775384 773948
NMB0742 conserved hypothetical protein 775684 776040
NMB0743 ubiquinone/menaquinone biosynthesis methylytransferase UbiE 776097
776831
NMB0744 hypothetical protein 777054 777530
NMB0745 2-amino-4-hydroxy-6-hydroxymethyldihydropteridine-
pyrophosphokinase 778153 777662
NMB0746 conserved hypothetical protein 778537 778166
NMB0747 conserved hypothetical protein 779157 778594
NMB0748 host factor-I 779535 779245
NMB0749 penicillin-binding protein 4 780602 779667
NMB0750 bacterioferritin comigratory protein 780923 781360
NMB0751 integrase/recombinase XerD 781415 782287
NMB0752 bacterioferritin-associated ferredoxin, putative 782462 782659
NMB0753 conserved hypothetical protein 782828 783058
NMB0754 hypothetical protein 783066 783173
NMB0755 hypothetical protein 783194 783334
NMB0756 dTDP-L-rhamnose synthase, putative 784398 783481
NMB0757 phosphoribosylaminoimidazole-succinocarboxamide synthase 784598
785458
NMB0758 polyribonucleotide nucleotidyltransferase 785695 787815
NMB0759 conserved hypothetical protein 788619 787894
NMB0760 diaminopimelate epimerase 789006 789854
NMB0761 hypothetical protein 789940 790164
NMB0762 hypothetical protein 790198 790653
NMB0763 cysteine synthase 790653 791582
NMB0764 conserved hypothetical protein 792048 792950
NMB0765 signal peptidase I 794128 793112
NMB0766 GTP-binding protein LepA 796064 794274
NMB0767 5-methylthioadenosine nucleosidase/S-adenosylhomocysteine
nucleosidase 796909 796211
NMB0768 twitching motility protein PilT 797095 798204
NMB0769 DNA polymerase III, delta prime subunit, putative 798241 799215
NMB0770 type IV pilus assembly protein PilZ, putative 799222 799569
NMB0771 conserved hypothetical protein 799577 800353
NMB0772 conserved hypothetical protein 800382 800594
NMB0773 conserved hypothetical protein 800698 801006
NMB0774 uracil phosphoribosyltransferase 801115 801738
NMB0775 hypothetical protein 801764 802081
NMB0776 conserved hypothetical protein 802335 802751
NMB0777 uroporphyrinogen-III synthase HemD, putative 802796 803533
NMB0778 uroporphyrin-III C-methyltransferase HemX, putative 803611 804882
NMB0779 hypothetical protein 804882 806102
NMB0780 hypothetical protein 806138 806575
NMB0781 uroporphyrinogen decarboxylase 806732 807793
NMB0782 DNA repair protein RadA 807982 809358
NMB0783 conserved hypothetical protein 810116 809640
NMB0784 phage shock protein E precursor, putative 810717 810361

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NMB0785 exodeoxyribonuclease V 135 KD polypeptide 814370 810759
NMB0786 conserved hypothetical protein 815358 814453
NMB0787 amino acid ABC transporter, periplasmic amino acid-binding protein
815643 816467
NMB0788 amino acid ABC transporter, permease protein 816514 817173
NMB0789 amino acid ABC transporter, ATP-binding protein 817186 817938
NMB0790 phosphoglucomutase 819343 817964
NMB0791 peptidyl-prolyl cis-trans isomerase 820019 819513
NMB0792 transporter, NadC family 821553 820141
NMB0793 hypothetical protein 821759 821553
NMB0794 hypothetical protein 822146 821787
NMB0795 peptidyl-tRNA hydrolase 822988 822413
NMB0796 conserved hypothetical protein 823319 823044
NMB0797 conserved hypothetical protein 823749 823315
NMB0798 cell division protein FtsH 825932 823968
NMB0799 cell division protein FtsJ 826616 825999
NMB0800 conserved hypothetical protein 826726 827007
NMB0801 delta-aminolevulinic acid dehydratase 827193 828191
NMB0802 cystathionine gamma-synthase 829414 828260
NMB0803 conserved hypothetical protein 829606 830376
NMB0804 NAD(P)H nitroreductase, putative 830489 831151
NMB0805 transposase, IS30 family 831295 832257
NMB0806 conserved hypothetical protein 833050 832295
NMB0807 conserved hypothetical protein 833965 833078
NMB0808 hypothetical protein 834551 833988
NMB0809 conserved hypothetical protein 835399 834605
NMB0810 transcriptional regulator, TetR family 836104 835457
NMB0811 UDP-N-acetylpyruvoylglucosamine reductase 837156 836119
NMB0812 conserved hypothetical protein 838579 837203
NMB0813 hypothetical protein 838634 838819
NMB0814 histidyl-tRNA synthetase 838914 840062
NMB0815 adenylosuccinate synthetase 840163 841464
NMB0816 hypothetical protein 841592 841903
NMB0817 hypothetical protein 841932 842312
NMB0818 hypothetical protein 842329 842736
NMB0819 hypothetical protein 842856 843245
NMB0820 hypothetical protein 843456 843845
NMB0821 hypothetical protein 843962 844519
NMB0822 heat shock protein HtpX 845866 844826
NMB0823 adenylate kinase 845878 846522
NMB0824 orotidine 5'-phosphate decarboxylase 847051 847788
NMB0825 ADP-heptose synthase, putative 847846 848814
NMB0826 C-5 cytosine-specific DNA methylase 848854 850086
NMB0827 type II restriction enzyme-related protein FRAMESHIFT 850091
851119
NMB0828 ADP-L-glycero-D-mannoheptose-6-epimerase 851251 852252
NMB0829 type I restriction enzyme EcoR124II M protein 852329 853870
NMB0830 conserved hypothetical protein 853870 854877
NMB0831 type I restriction enzyme S protein, degenerate 855046 856216
NMB0832 anticodon nuclease 856277 857416
NMB0833 type I restriction enzyme-related protein 857416 857799
NMB0834 transposase, IS30 family 858756 857794
NMB0835 type I restriction enzyme EcoR124II R protein, putative 858832
861594
NMB0836 ATP-dependent Clp protease, ATP-binding subunit ClpA 863945 861639
NMB0837 conserved hypothetical protein 864249 863950
NMB0838 cold-shock domain family protein 864492 864692
NMB0839 pmbA protein 866323 864995
NMB0840 conserved hypothetical protein 866446 866979
NMB0841 hypothetical protein 867029 867742
NMB0842 single-stranded-DNA-specific exonuclease RecJ 867814 869511
NMB0843 polyA polymerase 869811 871169
NMB0844 hypothetical protein 871345 871665
NMB0845 PhoH-related protein 872732 871782

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NMB0846 LPS biosynthesis protein-related protein 873905 872874
NMB0847 hypothetical protein 874235 874065
NMB0848 hypothetical protein 874369 875070
NMB0849 deoxycytidine triphosphate deaminase, putative 875703 875140
NMB0850 hypothetical protein 876185 875772
NMB0851 recombination associated protein RdgC 877146 876250
NMB0852 essential GTPase 878634 877180
NMB0853 conserved hypothetical protein 879413 878787
NMB0854 histidyl-tRNA synthetase 880709 879417
NMB0855 bacteriocin resistance protein, putative 881459 880806
NMB0856 hypothetical protein 882208 881744
NMB0857 hypothetical protein 882441 882268
NMB0858 hypothetical protein 882645 882448
NMB0859 hypothetical protein 883025 882651
NMB0860 hypothetical protein 883340 883086
NMB0861 hypothetical protein 883975 883433
NMB0862 hypothetical protein 884091 883975
NMB0863 hypothetical protein 884410 884141
NMB0864 hypothetical protein 884966 884679
NMB0865 hypothetical protein 885445 884975
NMB0866 hypothetical protein 886357 885491
NMB0867 YabO/YceC/SfhB family protein 886521 887441
NMB0868 conserved hypothetical protein 888163 887525
NMB0869 hypothetical protein 889009 888221
NMB0870 3-methyl-2-oxobutanoate hydroxymethyltransferase 889502 890290
NMB0871 pantoate--beta-alanine ligase 890416 891249
NMB0872 conserved hypothetical protein 891416 893257
NMB0873 outer membrane lipoprotein LolB, putative 893400 893978
NMB0874 conserved hypothetical protein 893991 894833
NMB0875 ribose-phosphate pyrophosphokinase 895258 896238
NMB0876 50S ribosomal protein L25 896308 896877
NMB0877 penicillin-binding protein 898174 897008
NMB0878 threonine dehydratase 898322 899845
NMB0879 sulfate ABC transporter, ATP-binding protein 900978 899908
NMB0880 sulfate ABC transporter, permease protein 901835 900978
NMB0881 sulfate ABC transporter, permease protein 902923 902090
NMB0882 hypothetical protein 903214 903543
NMB0883 conserved hypothetical protein 903878 904384
NMB0884 superoxide dismutase 905491 904907
NMB0885 replicative DNA helicase 905655 907058
NMB0886 fimbrial protein FimT 907370 908035
NMB0887 type IV pilus assembly protein PilV, putative 908056 908667
NMB0888 hypothetical protein 908667 909605
NMB0889 hypothetical protein 909587 910177
NMB0890 type IV pilin-related protein 910170 910655
NMB0891 hypothetical protein 911708 911944
NMB0892 AzlC-related protein 912795 912376
NMB0893 deoxyuridine 5'-triphosphate nucleotidohydrolase 912995 913444
NMB0894 aminotransferase, class I 913525 914709
NMB0895 conserved hypothetical protein 914975 915751
NMB0896 integrase, FRAMESHIFT 916283 917352
NMB0897 hypothetical protein 917468 917845
NMB0898 hypothetical protein 917894 918079
NMB0899 hypothetical protein 918396 918749
NMB0900 hypothetical protein 919621 920535
NMB0901 D-lactate dehydrogenase-related protein 920880 920599
NMB0902 hypothetical protein 921133 920945
NMB0903 hypothetical protein 921429 921139
NMB0904 hypothetical protein 921686 921429
NMB0905 hypothetical protein 921936 921724
NMB0906 hypothetical protein 922860 922009
NMB0907 hypothetical protein 923244 922888
NMB0908 hypothetical protein 923512 923315
NMB0909 hypothetical protein 924280 923759

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NMB0910 transcriptional regulator 925000 924287
NMB0911 transposase, IS30 family 926382 925420
NMB0912 hypothetical protein 926526 927149
NMB0913 pemK protein 927552 927208
NMB0914 pemI protein 927790 927557
NMB0915 hypothetical protein 928640 928152
NMB0916 hypothetical protein 928799 928662
NMB0917 death-on-curing protein 929446 929081
NMB0918 hypothetical protein 929574 929446
NMB0919 IS1106 transposase, putative 930929 929973
NMB0920 isocitrate dehydrogenase 934317 932095
NMB0921 hypothetical protein 934522 934325
NMB0922 alpha-2,3-sialyltransferase 934750 935862
NMB0923 cytochrome c 936488 936033
NMB0924 oxidoreductase, short-chain dehydrogenase/reductase family 936607
937425
NMB0925 acyl CoA thioester hydrolase family protein 937925 937482
NMB0926 opacity protein 940336 939513
NMB0927 proline iminopeptidase 941840 942769
NMB0928 hypothetical protein 944025 942832
NMB0929 dihydrodipicolinate synthase 944909 944037
NMB0930 xanthine/uracil permease family protein 945369 946757
NMB0931 RNA methyltransferase, TrmH family 947574 946825
NMB0932 conserved hypothetical protein 948129 947644
NMB0933 cytidine and deoxycytidylate deaminase family protein 948580
948137
NMB0934 DNA transformation protein tfoX-related protein 948853 948625
NMB0935 tRNA delta(2)-isopentenylpyrophosphate transferase 949798 948860
NMB0936 hypothetical protein 951481 950180
NMB0937 elongation factor P (EF-P) 951788 952345
NMB0938 hypothetical protein 953235 952402
NMB0939 conserved hypothetical protein 953933 953355
NMB0940 homoserine O-acetyltransferase 955069 953933
NMB0941 50S ribosomal protein L36 955756 955634
NMB0942 50S ribosomal protein L31, putative 956031 955759
NMB0943 5,10-methylenetetrahydrofolate reductase 956231 957106
NMB0944 5-methyltetrahydropteroyltriglutamate-homocysteine
methyltransferase 957247 959520
NMB0945 hypothetical protein 959535 959696
NMB0946 peroxiredoxin 2 family protein/glutaredoxin 959802 960536
NMB0947 lipoamide dehydrogenase, putative 960788 962188
NMB0948 succinate dehydrogenase, cytochrome b556 subunit 962470 962844
NMB0949 succinate dehydrogenase, hydrophobic membrane anchor protein
962841 963179
NMB0950 succinate dehydrogenase, flavoprotein subunit 963185 964945
NMB0951 succinate dehydrogenase, iron-sulfur protein 965068 965772
NMB0952 conserved hypothetical protein 965779 966024
NMB0953 hypothetical protein 966024 966104
NMB0954 citrate synthase 966139 967419
NMB0955 2-oxoglutarate dehydrogenase, E1 component 967627 970452
NMB0956 2-oxoglutarate dehydrogenase, E2 component, dihydrolipoamide
succinyltransferase 970555 971733
NMB0957 2-oxoglutarate dehydrogenase, E3 component, lipoamide
dehydrogenase 972101 973531
NMB0958 hypothetical protein 973659 973943
NMB0959 succinyl-CoA synthetase, beta subunit 974045 975208
NMB0960 succinyl-CoA synthetase, alpha subunit 975222 976109
NMB0961 funZ protein 978267 976675
NMB0962 excinuclease ABC, subunit A 981150 978304
NMB0963 phosphatidylserine decarboxylase precursor-related protein 981305
982099
NMB0964 TonB-dependent receptor 985503 983230
NMB0965 hypothetical protein 985832 985564

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NMB0966 para-aminobenzoate synthase glutamine amidotransferase component
II 985925 986512

NMB0967 anthranilate phosphoribosyltransferase 986579 987634

NMB0968 hypothetical protein 987644 987729

NMB0969 hypothetical protein 988030 987792

NMB0970 conserved hypothetical protein, FRAMESHIFT 988106 989527

NMB0971 hypothetical protein 989493 989780

NMB0972 hypothetical protein 989788 989982

NMB0973 hypothetical protein 989993 990274

NMB0974 hypothetical protein 990284 990559

NMB0975 hypothetical protein 990690 991004

NMB0976 TspB-related protein 990991 991383

NMB0977 modulator of drug activity B, putative 991676 992146

NMB0978 NAD(P) transhydrogenase, beta subunit 993742 992360

NMB0979 hypothetical protein 994205 993825

NMB0980 NAD(P) transhydrogenase, alpha subunit 995750 994212

NMB0981 phosphoserine phosphatase 996040 996870

NMB0982 chloride channel protein-related protein 997018 998157

NMB0983 phosphoribosylaminoimidazolecarboxamide formyltransferase/IMP
cyclohydrolase 998324 999901

NMB0984 transposase, putative, degenerate 1000517 1001457

NMB0985 E16-related protein 1001522 1002016

NMB0986 hypothetical protein 1001997 1002425

NMB0987 N-acetylmuramoyl-L-alanine amidase, putative 1002736 1003278

NMB0988 hypothetical protein 1003278 1003478

NMB0989 hypothetical protein 1003484 1003645

NMB0990 hypothetical protein 1003859 1004260

NMB0991 IS1106 transposase 1005417 1004308

NMB0992 adhesin 1007326 1005554

NMB0993 rubredoxin 1009428 1009261

NMB0994 acyl-CoA dehydrogenase family protein 1011202 1010114

NMB0995 macrophage infectivity potentiator-related protein 1012020 1011340

NMB0996 hypothetical protein 1012411 1012043

NMB0997 D-lactate dehydrogenase 1014397 1012709

NMB0998 oxidoreductase, putative 1014921 1018751

NMB0999 Nifr3/SMM1 family protein 1018935 1019933

NMB1000 IS1106 transposase, putative FRAMESHIFT 1020537 1021551

NMB1001 integrase protein, degenerate 1023183 1022614

NMB1002 hypothetical protein 1024370 1023498

NMB1003 hypothetical protein 1024711 1024418

NMB1004 hypothetical protein 1024962 1024720

NMB1005 hypothetical protein 1025179 1024958

NMB1006 hypothetical protein 1025360 1025184

NMB1007 transcriptional regulator 1025451 1025819

NMB1008 hypothetical protein 1025824 1026444

NMB1009 conserved hypothetical protein 1026440 1026631

NMB1010 hypothetical protein 1026658 1027218

NMB1011 hypothetical protein 1027252 1028196

NMB1012 hypothetical protein 1028284 1028784

NMB1013 hypothetical protein 1028801 1028971

NMB1014 conserved hypothetical protein 1029045 1029635

NMB1015 IS150 transposase, putative FRAMESHIFT 1029653 1030443

NMB1016 conserved hypothetical protein 1031794 1031192

NMB1017 sulfate ABC transporter, periplasmic sulfate-binding protein
1033574 1032522

NMB1018 conserved hypothetical protein 1034162 1033683

NMB1019 phosphoribosylaminoimidazole carboxylase, ATPase subunit 1035345
1034212

NMB1020 hypothetical protein 1035887 1035345

NMB1021 anthranilate synthase component I 1037359 1035887

NMB1022 transposase, IS30 family 1038444 1037482

NMB1023 conserved hypothetical protein 1039543 1038587

NMB1024 conserved hypothetical protein 1040502 1039639

NMB1025 conserved hypothetical protein 1040896 1040537

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NMB1026 conserved hypothetical protein 1040971 1041447
NMB1027 dnaJ protein, truncation 1041473 1042192
NMB1028 conserved hypothetical protein 1042197 1043069
NMB1029 aspartate ammonia-lyase 1044541 1043147
NMB1030 conserved hypothetical protein 1045565 1045005
NMB1031 3-isopropylmalate dehydrogenase 1046798 1045731
NMB1032 type II restriction enzyme NlaIV 1047563 1046835
NMB1033 modification methylase NlaIV 1048850 1047582
NMB1034 3-isopropylmalate dehydratase, small subunit 1049666 1049028
NMB1035 hypothetical protein 1049982 1049731
NMB1036 3-isopropylmalate dehydratase, large subunit 1051488 1050082
NMB1037 glutamate--cysteine ligase 1051748 1053094
NMB1038 DNA repair protein RadC 1053220 1053894
NMB1039 conserved hypothetical protein 1053970 1054692
NMB1040 hypothetical protein 1054848 1056125
NMB1041 GTP-binding protein 1056133 1057308
NMB1042 cation transport ATPase, E1-E2 family 1057308 1059776
NMB1043 hypothetical protein 1059940 1060142
NMB1044 ferredoxin--NADP reductase 1061316 1060543
NMB1045 hypothetical protein 1062298 1061507
NMB1046 threonine synthase 1063753 1062347
NMB1047 hypothetical protein 1064197 1063829
NMB1048 hypothetical protein 1065918 1064452
NMB1049 transcriptional regulator, putative 1066174 1067085
NMB1050 transposase, IS30 family 1068512 1067550
NMB1051 ABC transporter, ATP-binding protein 1070544 1068637
NMB1052 dedA protein 1071207 1070566
NMB1053 class 5 outer membrane protein 1072189 1071374
NMB1054 IS1106 transposase, degenerate 1073920 1072988
NMB1055 serine hydroxymethyltransferase 1075474 1074227
NMB1056 hypothetical protein 1075753 1075544
NMB1057 gamma-glutamyltranspeptidase 1077776 1075959
NMB1058 conserved hypothetical protein FRAMESHIFT 1078161 1077902
NMB1059 conserved hypothetical protein 1078505 1078720
NMB1060 fructose-1,6-bisphosphatase 1079840 1078869
NMB1061 conserved hypothetical protein 1080931 1080089
NMB1062 conserved hypothetical protein 1081610 1081011
NMB1063 dihydroneopterin aldolase 1081666 1082019
NMB1064 conserved hypothetical protein 1082056 1082589
NMB1065 crcB protein 1083465 1083109
NMB1066 hypothetical protein 1084174 1083497
NMB1067 cell division protein FtsK 1084339 1087380
NMB1068 gamma-glutamyl phosphate reductase 1088870 1087611
NMB1069 glutamate 5-kinase 1089992 1088886
NMB1070 2-isopropylmalate synthase 1090477 1092027
NMB1071 conserved hypothetical protein 1092125 1092784
NMB1072 prolipoprotein diacylglycerol transferase 1093721 1092873
NMB1073 conserved hypothetical protein 1094922 1093795
NMB1074 acetylglutamate kinase 1095092 1095985
NMB1075 conserved hypothetical protein 1098302 1097637
NMB1076 DnaA-related protein 1098967 1098302
NMB1077 ABC transporter, ATP-binding protein, truncation 1099623 1099075
NMB1078 transcriptional regulator, UmuD/LexA family 1100312 1099875
NMB1079 hypothetical protein 1100580 1100425
NMB1080 ner protein FRAMESHIFT 1100802 1101061
NMB1081 bacteriophage transposase 1101126 1103108
NMB1082 hypothetical protein 1103120 1103317
NMB1083 bacteriophage DNA transposition protein B, putative 1103481
1104650
NMB1084 hypothetical protein 1104655 1105173
NMB1085 N-acetylmuramoyl-L-alanine amidase, putative 1105319 1105861
NMB1086 hypothetical protein 1106234 1106467
NMB1087 hypothetical protein 1106758 1107060
NMB1088 conserved hypothetical protein 1107278 1107111

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NMB1089 hypothetical protein 1107506 1107841
NMB1090 hypothetical protein 1107856 1108119
NMB1091 hypothetical protein 1108119 1108313
NMB1092 hypothetical protein 1108319 1108822
NMB1093 hypothetical protein 1109412 1108825
NMB1094 hypothetical protein 1109497 1111044
NMB1095 conserved hypothetical protein 1111047 1112612
NMB1096 conserved hypothetical protein 1112602 1113894
NMB1097 cryptic Mu-phage G protein, putative 1114007 1114419
NMB1098 I protein, putative 1114653 1115711
NMB1099 transposase, IS30 family 1116767 1115805
NMB1100 hypothetical protein 1116795 1117274
NMB1101 conserved hypothetical protein 1117277 1117696
NMB1102 hypothetical protein 1117746 1118336
NMB1103 hypothetical protein 1118336 1118530
NMB1104 phage sheath protein 1118536 1119942
NMB1105 hypothetical protein 1120010 1120384
NMB1106 hypothetical protein 1120391 1120753
NMB1107 hypothetical protein 1121610 1121011
NMB1108 hypothetical protein 1121780 1123933
NMB1109 phage virion protein, putative 1123936 1125264
NMB1110 tail protein, 43 kDa 1125257 1126399
NMB1111 baseplate assembly protein V, putative 1126399 1127064
NMB1112 conserved hypothetical protein 1127168 1127512
NMB1113 conserved hypothetical protein FRAMESHIFT 1127529 1128580
NMB1114 conserved hypothetical protein 1128580 1129137
NMB1115 tail fibre protein, putative 1129151 1131121
NMB1116 hypothetical protein 1131560 1132084
NMB1117 hypothetical protein 1132350 1132204
NMB1118 conserved hypothetical protein 1132762 1132478
NMB1119 conserved hypothetical protein 1132842 1133444
NMB1120 hypothetical protein 1133426 1133719
NMB1121 conserved hypothetical protein 1133719 1133925
NMB1122 ABC transporter, ATP-binding protein FRAMESHIFT 1135181 1134041
NMB1198 conserved hypothetical protein 1199352 1198465
NMB1161 hypothetical protein 1167620 1167426
NMB1162 hypothetical protein 1168307 1167663
NMB1163 hypothetical protein 1168675 1168307
NMB1164 hypothetical protein 1169353 1168685
NMB1165 oxidoreductase, short chain dehydrogenase/reductase family 1170237
1169521
NMB1128 conserved hypothetical protein 1139597 1138287
NMB1167 hypothetical protein 1171869 1171666
NMB1168 phytoene synthase, putative 1172903 1172034
NMB1131 chaperone protein HscA 1142897 1141038
NMB1132 hypothetical protein 1143630 1142977
NMB1171 conserved hypothetical protein / ankyrin-related protein 1176464
1175706
NMB1172 ferredoxin, 2Fe-2S type 1176860 1176522
NMB1173 hypothetical protein 1177278 1177138
NMB1136 hypothetical protein 1146017 1145337
NMB1175 conserved hypothetical protein 1178247 1178053
NMB1176 conserved hypothetical protein 1178719 1178321
NMB1139 acetyl-CoA carboxylase, carboxyl transferase alpha subunit 1147851
1146895
NMB1140 mesJ protein FRAMESHIFT 1149229 1147948
NMB1179 RNA methyltransferase, TrmH family 1182124 1181516
NMB1180 hypothetical protein 1182411 1182178
NMB1181 hypothetical protein 1182945 1182583
NMB1182 hypothetical protein 1183262 1182960
NMB1145 UDP-N-acetylmuramate:L-alanyl-gamma-D-glutamyl-meso-
diaminopimelate ligase 1152664 1151291
NMB1146 biotin synthetase 1153923 1152874
NMB1185 hypothetical protein 1186675 1186043

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NMB1148 hypothetical protein 1154845 1154693
NMB1187 hypothetical protein 1187052 1186912
NMB1150 dihydroxy-acid dehydratase 1157144 1155288
NMB1189 sulfite reductase hemoprotein, beta-component 1191122 1189356
NMB1190 sulfite reductase (NADPH) flavoprotein, alpha component 1192963
1191152
NMB1153 sulfate adenylyltransferase, subunit 1 1162210 1160927, plasmid
protein [REDACTED]
NMB1192 sulfate adenylyltransferase, subunit 2 1195208 1194288
NMB1155 phosphoadenosine phosphosulfate reductase 1163950 1163213
NMB1194 siroheme synthase 1197448 1196000
NMB1195 hypothetical protein 1197732 1197577
NMB1158 nickel-dependent hydrogenase, b-type cytochrome subunit 1166365
1165712
NMB1197 conserved hypothetical protein 1199352 1198465
NMB1199 GTP-binding protein TypA 1201433 1199625
NMB1200 ribonuclease II family protein 1202272 1204644
NMB1201 IMP dehydrogenase 1206449 1204989
NMB1202 hypothetical protein 1207237 1206779
NMB1203 protein-PII uridylyltransferase 1209886 1207331
NMB1204 transcriptional regulator 1210255 1209938
NMB1205 hypothetical protein 1210426 1210283
NMB1206 bacterioferritin B 1211053 1210583
NMB1207 bacterioferritin A 1211545 1211084
NMB1208 hypothetical protein 1211610 1211810
NMB1209 hypothetical protein 1211900 1212100
NMB1210 toxin-activating protein, putative 1212121 1212585
NMB1211 hypothetical protein 1212984 1212745
NMB1212 hypothetical protein 1213319 1212984
NMB1213 hypothetical protein 1213678 1213319
NMB1214 hemagglutinin/hemolysin-related protein 1220496 1213678
NMB1215 hypothetical protein 1220814 1220659
NMB1216 lipoic acid synthetase 1221989 1221009
NMB1217 lipoate-protein ligase B 1222554 1221985
NMB1218 conserved hypothetical protein 1222882 1222610
NMB1219 transporter, putative 1223067 1224134
NMB1220 stomatin/Mec-2 family protein 1225281 1224337
NMB1221 hypothetical protein 1225703 1225299
NMB1222 uracil-DNA glycosylase 1225784 1226440
NMB1223 site-specific DNA methylase, degenerate 1226520 1229028
NMB1224 hypothetical protein 1229552 1229154
NMB1225 hypothetical protein 1230112 1229600
NMB1226 ABC transporter, ATP-binding protein 1232500 1230581
NMB1227 conserved hypothetical protein 1232972 1232580
NMB1228 homoserine dehydrogenase 1233145 1234449
NMB1229 hypothetical protein 1234445 1234876
NMB1230 DNA-binding protein HU-beta 1235207 1234941
NMB1231 ATP-dependent protease La 1237851 1235392
NMB1232 conserved hypothetical protein 1238285 1239202
NMB1233 exodeoxyribonuclease V, alpha subunit 1240978 1239236
NMB1234 ABC transporter, ATP-binding protein 1241741 1241049
NMB1235 conserved hypothetical protein 1242981 1241737
NMB1236 hypothetical protein 1243186 1243461
NMB1237 recombination protein RecR 1244140 1243523
NMB1238 peptidyl-prolyl cis-trans isomerase-related protein 1245742
1244207
NMB1239 conserved hypothetical protein 1246176 1245805
NMB1240 ABC transporter, ATP-binding protein 1246326 1247951
NMB1241 tRNA nucleotidyltransferase 1248026 1249276
NMB1242 hypothetical protein 1249502 1249807
NMB1243 Holliday junction DNA helicase RuvB 1249892 1250920
NMB1244 ribulose-phosphate 3-epimerase 1251674 1250949
NMB1245 hypothetical protein 1252367 1252035
NMB1246 conserved hypothetical protein 1253294 1252434

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NMB1247 riboflavin synthase, alpha subunit 1254006 1253305
NMB1248 molybdopterin-guanine dinucleotide biosynthesis protein A
FRAMESHIFT 1254659 1254085
NMB1249 nitrate/nitrite sensory protein NarX, putative 1254901 1256670
NMB1250 transcriptional regulator, LuxR family 1256670 1257323
NMB1251 transposase, IS30 family 1258731 1257769
NMB1252 phosphoribosylformylglycinamide cyclo-ligase 1259914 1258883
NMB1253 hypothetical protein 1260672 1261346
NMB1254 GTP cyclohydrolase II 1261342 1261932
NMB1255 glycosyl transferase, degenerate 1262256 1263263
NMB1256 GTP cyclohydrolase II/3,4-dihydroxy-2-butanone-4-phosphate
synthase 1263728 1264816
NMB1257 site-specific DNA methylase, degenerate 1265357 1265130
NMB1258 conserved hypothetical protein 1267046 1265739
NMB1259 transposase, IS30 family 1267584 1268546
NMB1260 type III restriction-modification system EcoPI enzyme, subunit res
1271565 1268629
NMB1261 type III restriction-modification system EcoPI enzyme, subunit mod
POINT MUTATION FRAMESHIFT 1273661 1271581
NMB1262 peptidyl-prolyl cis-trans isomerase 1274334 1273780
NMB1263 CobW-related protein 1275316 1274402
NMB1264 conserved hypothetical protein 1275771 1275502
NMB1265 conserved hypothetical protein 1276061 1275771
NMB1266 zinc uptake regulation protein, putative 1276582 1276109
NMB1267 low molecular weight protein tyrosine-phosphatase 1277108 1276656
NMB1268 conserved hypothetical protein 1278348 1277236
NMB1269 hypothetical protein 1279559 1278465
NMB1270 conserved hypothetical protein 1281272 1279644
NMB1271 mercury transport periplasmic protein, putative 1281584 1281375
NMB1272 hypothetical protein 1281765 1281625
NMB1273 alginate O-acetylation protein AlgI, putative 1282215 1283648
NMB1274 hypothetical protein 1283662 1284642
NMB1275 hypothetical protein 1284642 1286083
NMB1276 long-chain-fatty-acid--CoA ligase 1286122 1287672
NMB1277 transporter, BCCT family 1289792 1287768
NMB1278 site-specific recombinase 1290081 1292084
NMB1279 membrane-bound lytic murein transglycosylase B, putative 1293319
1292213
NMB1280 very long chain acyl-CoA dehydrogenase-related protein 1294948
1293524
NMB1281 transcription-repair coupling factor 1295133 1299269
NMB1282 aspartate 1-decarboxylase 1299421 1299801
NMB1283 2-dehydro-3-deoxyphosphooctonate aldolase 1299826 1300665
NMB1284 hypothetical protein 1300683 1301120
NMB1285 enolase 1301171 1302454
NMB1286 conserved hypothetical protein 1302471 1302746
NMB1287 ferredoxin, putative 1303080 1302793
NMB1288 ribonucleoside-diphosphate reductase, beta subunit 1304479 1303328
NMB1289 type II restriction enzyme, putative 1305706 1304522
NMB1290 C-5 cytosine-specific DNA-methylase 1306712 1305702
NMB1291 ribonucleoside-diphosphate reductase, alpha subunit 1309049
1306773
NMB1292 hypothetical protein 1309394 1309209
NMB1293 hypothetical protein 1309563 1309886
NMB1294 1-acyl-sn-glycerol-3-phosphate acyltransferase 1310967 1310203
NMB1295 formamidopyrimidine-DNA glycosylase 1311882 1311058
NMB1296 hypothetical protein 1312599 1311937
NMB1297 membrane-bound lytic murein transglycosylase D 1312778 1314751
NMB1298 ribosomal small subunit pseudouridine synthase A 1314822 1315511
NMB1299 sodium- and chloride-dependent transporter, degenerate 1316091
1317454
NMB1300 cytidylate kinase 1317701 1318354
NMB1301 30S ribosomal protein S1 1318513 1320195
NMB1302 integration host factor, beta subunit 1320209 1320520

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NMB1303 transcriptional regulator, MerR family 1321281 1320877
NMB1304 alcohol dehydrogenase, class III 1321402 1322535
NMB1305 esterase, putative 1322547 1323371
NMB1306 conserved hypothetical protein 1323765 1324913
NMB1307 nucleoside diphosphate kinase 1324975 1325397
NMB1308 conserved hypothetical protein 1325543 1326634
NMB1309 fimbrial biogenesis and twitching motility protein, putative, 1326640 1327398
NMB1310 gcpE protein 1327417 1328679
NMB1311 hypothetical protein 1328970 1328737
NMB1312 ATP-dependent Clp protease, proteolytic subunit 1329655 1329128
NMB1313 trigger factor 1331148 1329838
NMB1314 cell division protein FtsK 1333791 1331356
NMB1315 uracil permease 1334014 1335222
NMB1316 hypothetical protein 1335289 1335726
NMB1317 hypothetical protein 1335865 1336266
NMB1318 CDP-diacylglycerol--serine O-phosphatidyltransferase 1336343 1337086
NMB1319 conserved hypothetical protein 1337090 1337860
NMB1320 50S ribosomal protein L9 1338540 1338091
NMB1321 30S ribosomal protein S18 1338787 1338560
NMB1322 primosomal replication protein n, putative 1339096 1338797
NMB1323 30S ribosomal protein S6 1339465 1339100
NMB1324 thioredoxin reductase 1340571 1339624
NMB1325 cation transport ATPase, E1-E2 family 1340710 1342869
NMB1326 excinuclease ABC, subunit C 1342969 1344819
NMB1327 conserved hypothetical protein 1345045 1346445
NMB1328 conserved hypothetical protein 1346570 1347283
NMB1329 hypothetical protein 1347649 1347840
NMB1330 hypothetical protein 1348276 1347917
NMB1331 excinuclease ABC, subunit B 1350416 1348392
NMB1332 carboxy-terminal peptidase 1352229 1350748
NMB1333 conserved hypothetical protein 1354146 1352359
NMB1334 hypothetical protein 1354238 1354471
NMB1335 creA protein 1354474 1355031
NMB1336 conserved hypothetical protein 1355036 1355581
NMB1337 conserved hypothetical protein 1355577 1356029
NMB1338 isomerase, putative 1356698 1356045
NMB1339 prolyl-tRNA synthetase 1358473 1356764
NMB1340 hypothetical protein 1358924 1359151
NMB1341 pyruvate dehydrogenase, E1 component 1359167 1361827
NMB1342 pyruvate dehydrogenase, E2 component, dihydrolipoamide acetyltransferase FRAMESHIFT 1361979 1363583
NMB1343 hypothetical protein 1363680 1364114
NMB1344 pyruvate dehydrogenase, E3 component, lipoamide dehydrogenase 1364135 1365916
NMB1345 hypothetical protein 1367830 1366283
NMB1346 TonB-dependent receptor, putative FRAMESHIFT 1369731 1367957
NMB1347 extragenic suppressor protein SuhB 1370786 1370004
NMB1348 RNA methylase, putative 1371030 1371842
NMB1349 hypothetical protein 1371906 1372760
NMB1350 hypothetical protein 1372967 1373305
NMB1351 fmu and fmv protein, putative 1373656 1374909
NMB1352 hypothetical protein 1375272 1375703
NMB1353 aldehyde dehydrogenase family protein 1377097 1375757
NMB1354 conserved hypothetical protein 1377755 1377105
NMB1355 glutamyl-tRNA (Gln) amidotransferase subunit C, putative 1377906 1378193
NMB1356 Glu-tRNA(Gln) amidotransferase, subunit A 1378259 1379701
NMB1357 conserved hypothetical protein 1379701 1380630
NMB1358 Glu-tRNA(Gln) amidotransferase, subunit B 1380676 1382103
NMB1359 CDP-6-deoxy-delta-3,4-glucose reductase, putative 1382318 1383325
NMB1360 pyridoxamine 5-phosphate oxidase 1384090 1383461

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NMB1361 conserved hypothetical protein 1384312 1385361
NMB1362 oxalate/formate antiporter, putative 1386974 1385436
NMB1363 exodeoxyribonuclease, large subunit 1388622 1387270
NMB1364 NH(3)-dependent NAD⁺ synthetase NadE, putative 1388819 1389637
NMB1365 conserved hypothetical protein 1390183 1389713
NMB1366 thioredoxin 1390481 1390810
NMB1367 conserved hypothetical protein 1391930 1390869
NMB1368 ATP-dependent RNA helicase, putative 1392141 1393526
NMB1369 hypothetical protein 1394572 1394021
NMB1370 hypothetical protein 1395217 1394860
NMB1371 acetylornithine aminotransferase 1395561 1396754
NMB1372 ATP-dependent Clp protease, ATP-binding subunit ClpX 1398104
1396863
NMB1373 ribosome-binding factor A 1398295 1398663
NMB1374 tRNA pseudouridine synthase B 1398699 1399619
NMB1375 modification methylase, putative FRAMESHIFT 1399839 1401945
NMB1376 conserved hypothetical protein POINT MUTATION 1401938 1404712
NMB1377 L-lactate dehydrogenase 1406036 1404867
NMB1378 conserved hypothetical protein 1406327 1406770
NMB1379 nifS protein 1406802 1408013
NMB1380 nifU protein 1408280 1408663
NMB1381 HesB/YadR/YfhF family protein 1408693 1409070
NMB1382 conserved hypothetical protein 1409254 1409036
NMB1383 chaperone protein HscB 1409336 1409833
NMB1384 DNA gyrase subunit A 1409934 1412681
NMB1385 IS1016 family transposase, degenerate 1412841 1413241
NMB1386 transposase, putative FRAMESHIFT 1413303 1413955
NMB1387 hypothetical protein 1414840 1414292
NMB1388 glucose-6-phosphate isomerase 1416500 1414857
NMB1389 RpiR/YebK/YfhH family protein 1417469 1416624
NMB1390 glucokinase 1418505 1417522
NMB1391 oxidoreductase, Sol/DevB family 1419181 1418489
NMB1392 glucose-6-phosphate 1-dehydrogenase 1420906 1419464
NMB1393 phosphogluconate dehydratase 1421474 1423306
NMB1394 4-hydroxy-2-oxoglutarate aldolase/2-dehydro-3-deoxyphosphogluconate
aldolase 1423490 1424125
NMB1395 alcohol dehydrogenase, zinc-containing 1425427 1424390
NMB1396 A/G-specific adenine glycosylase 1425581 1426627
NMB1397 hypothetical protein 1426793 1426972
NMB1398 Cu-Zn-superoxide dismutase 1427047 1427604
NMB1399 IS1106 transposase 1429146 1428175
NMB1400 ABC transporter family protein 1431631 1429406
NMB1401 IS1016C2 transposase 1432983 1432447
NMB1402 hypothetical protein 1433320 1433751
NMB1403 FrpA/C-related protein 1433795 1433983
NMB1404 hypothetical protein 1434021 1434746
NMB1405 FrpA/C-related protein 1434763 1435962
NMB1406 hypothetical protein 1436396 1436755
NMB1407 FrpA-related protein, degenerate 1436755 1437881
NMB1408 hypothetical protein 1437960 1438451
NMB1409 FrpA/C-related protein 1438582 1439007
NMB1410 hypothetical protein 1439247 1439783
NMB1411 IS1016C2 transposase 1440610 1439960
NMB1412 FrpC operon protein 1441216 1442022
NMB1413 IS1016 family transposase, putative FRAMESHIFT 1442715 1442132
NMB1414 FrpC operon protein 1442798 1443568
NMB1415 iron-regulated protein FrpC 1443588 1449074
NMB1416 aminopeptidase N 1452022 1449422
NMB1417 conserved hypothetical protein 1452947 1452156
NMB1418 HtrB/MsbB family protein 1454563 1453697
NMB1419 crossover junction endodeoxyribonuclease RuvC 1455150 1454617
NMB1420 factor-for-inversion stimulation protein Fis, putative 1455392
1455156
NMB1421 nifR3 protein 1456432 1455425

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NMB1422 ATP-dependent RNA helicase, putative 1456798 1458168
NMB1423 conserved hypothetical protein 1458746 1459870
NMB1424 hypothetical protein 1459903 1460928
NMB1425 lysyl-tRNA synthetase, heat inducible 1462560 1461052
NMB1426 hypothetical protein 1463968 1462718
NMB1427 hypothetical protein 1464208 1464032
NMB1428 aminopeptidase, putative 1464426 1466219
NMB1429 outer membrane protein PorA 1468209 1467034
NMB1430 transcription elongation factor GreA 1470964 1470491
NMB1431 hypothetical protein 1471298 1471050
NMB1432 3-phosphoshikimate 1-carboxyvinyltransferase 1471360 1472658
NMB1433 conserved hypothetical protein FRAMESHIFT 1473237 1472707
NMB1434 cardiolipin synthetase family protein 1474971 1473448
NMB1435 drug resistance translocase family protein 1476489 1475086
NMB1436 conserved hypothetical protein 1476774 1477550
NMB1437 conserved hypothetical protein 1477550 1478248
NMB1438 conserved hypothetical protein 1478248 1479699
NMB1439 phosphoribosylaminoimidazole carboxylase, catalytic subunit
1480370 1479888
NMB1440 hypothetical protein 1481131 1480421
NMB1441 O-methyltransferase, putative 1481799 1481134
NMB1442 mismatch repair protein MutL 1482139 1484112
NMB1443 DNA polymerase III, subunits gamma and tau 1484210 1486321
NMB1444 conserved hypothetical protein 1486404 1486736
NMB1445 recA protein 1489556 1488513
NMB1446 3-dehydroquinase dehydratase 1489810 1490571
NMB1447 ATP-dependent DNA helicase Rep 1490594 1492606
NMB1448 DNA-damage-inducible protein P 1493734 1492781
NMB1449 TonB-dependent receptor POINT MUTATION 1496967 1493881
NMB1450 ferredoxin--NADP reductase 1497241 1498017
NMB1451 DNA polymerase III, epsilon subunit 1499643 1498234
NMB1452 conserved hypothetical protein 1500459 1501595
NMB1453 hypothetical protein 1502335 1501847
NMB1454 ferredoxin, 4Fe-4S bacterial type 1503891 1502398
NMB1455 hypothetical protein 1504075 1503959
NMB1456 hypothetical protein 1504347 1504153
NMB1457 transketolase 1504419 1506395
NMB1458 fumarate hydratase, class II 1506547 1507932
NMB1459 conserved hypothetical protein 1508923 1508003
NMB1460 single-strand binding protein 1509972 1509451
NMB1461 drug resistance translocase family protein 1511361 1509979
NMB1462 transglycosylase, putative 1512092 1511472
NMB1463 IS1106 transposase, degenerate 1512998 1512596
NMB1464 conserved hypothetical protein 1513541 1513053
NMB1465 opacity protein FRAMESHIFT 1515309 1514483
NMB1466 conserved hypothetical protein 1515639 1516367
NMB1467 exopolyphosphatase 1516487 1517992
NMB1468 hypothetical protein 1518527 1518207
NMB1469 hypothetical protein 1518607 1518527
NMB1470 hypothetical protein 1519392 1518850
NMB1471 tryptophanyl-tRNA synthetase 1520471 1519464
NMB1472 clpB protein 1520732 1523308
NMB1473 aminotransferase, class I 1524612 1523401
NMB1474 4-oxalocrotonate tautomerase, putative 1524910 1524704
NMB1475 conserved hypothetical protein 1525255 1526058
NMB1476 glutamate dehydrogenase, NAD-specific 1527384 1526122
NMB1477 hypothetical protein 1527562 1527396
NMB1478 phosphoglycolate phosphatase FRAMESHIFT 1527786 1528489
NMB1479 regulatory protein RecX 1528560 1529018
NMB1480 hypothetical protein 1529095 1529253
NMB1481 hypothetical protein 1529262 1529393
NMB1482 acyl CoA thioester hydrolase family protein 1529409 1529888
NMB1483 lipoprotein NlpD, putative 1531499 1530255
NMB1484 stationary-phase survival protein SurE 1532501 1531758

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NMB1485 conserved hypothetical protein 1534074 1532521
NMB1486 hypothetical protein 1534263 1534126
NMB1487 fimbrial assembly protein 1535230 1534445
NMB1488 succinate-semialdehyde dehydrogenase (NADP+) 1536772 1535342
NMB1489 hypothetical protein 1537259 1537750
NMB1490 hypothetical protein 1538345 1537917
NMB1491 hypothetical protein 1538785 1538699
NMB1492 hypothetical protein 1538860 1538795
NMB1493 carbon starvation protein A 1538892 1540970
NMB1494 conserved hypothetical protein 1540963 1541154
NMB1495 hypothetical protein 1541371 1541562
NMB1496 conserved hypothetical protein 1541673 1542230
NMB1497 TonB-dependent receptor 1543234 1545996
NMB1498 aspartokinase, alpha and beta subunits 1549220 1548006
NMB1499 ribonuclease PH 1550148 1549423
NMB1500 conserved hypothetical protein 1550694 1550233
NMB1501 HesA/MoeB/ThiF family protein 1550911 1551684
NMB1502 hypothetical protein 1551825 1552349
NMB1503 hypothetical protein 1552608 1552814
NMB1504 conserved hypothetical protein 1552706 1553557
NMB1505 nicotinate phosphoribosyltransferase 1553601 1554806
NMB1506 arginyl-tRNA synthetase 1554901 1556616
NMB1507 hypothetical protein 1556714 1557070
NMB1508 hypothetical protein 1557130 1558584
NMB1509 amino acid ABC transporter, permease protein 1560344 1559601
NMB1510 thermonuclease family protein 1561224 1560526
NMB1511 ribose 5-phosphate isomerase A 1561934 1561266
NMB1512 YgbB/YacN family protein 1562493 1562014
NMB1513 conserved hypothetical protein 1563214 1562528
NMB1514 DNA polymerase III, epsilon subunit 1563945 1563214
NMB1515 transporter, putative 1565411 1564104
NMB1516 fixS protein 1565589 1565404
NMB1517 hypothetical protein 1565885 1565589
NMB1518 acetate kinase 1566236 1567429
NMB1519 thiol:disulfide interchange protein DsbD 1569752 1567950
NMB1520 hypothetical protein 1570337 1569819
NMB1521 phytoene synthase-related protein 1571249 1570425
NMB1522 FKBP-type peptidyl-prolyl cis-trans isomerase SlyD 1571803 1571324
NMB1523 hypothetical protein 1572276 1572569
NMB1524 oxidoreductase, putative 1572682 1574046
NMB1525 VirG-related protein FRAMESHIFT 1576262 1574233
NMB1526 small major protein B 1577081 1576638
NMB1527 ADP-heptose--LPS heptosyltransferase II 1578146 1577139
NMB1528 methylated-DNA--protein-cysteine methyltransferase, putative
1579353 1578547
NMB1529 conserved hypothetical protein FRAMESHIFT 1579597 1580409
NMB1530 succinyl-diaminopimelate desuccinylase 1582228 1581086
NMB1531 conserved hypothetical protein 1582961 1582344
NMB1532 conserved hypothetical protein 1583504 1582998
NMB1533 H.8 outer membrane protein 1584150 1583602
NMB1534 hypothetical protein 1584287 1584150
NMB1535 hypothetical protein 1584404 1584874
NMB1536 preprotein translocase SecA subunit 1584984 1587731
NMB1537 DNA primase 1587879 1589648
NMB1538 RNA polymerase sigma factor RpoD 1589838 1591763
NMB1539 IS1106 transposase 1591913 1592917
NMB1540 lactoferrin-binding protein A 1597271 1594443
NMB1541 lactoferrin-binding protein B 1599481 1597271
NMB1542 hypothetical protein 1600504 1600722
NMB1543 conserved hypothetical protein 1600871 1602082
NMB1544 hypothetical protein 1602097 1602405
NMB1545 hypothetical protein 1602412 1602609
NMB1546 hypothetical protein 1602795 1603076
NMB1547 hypothetical protein 1603107 1603406

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NMB1548 tspB protein, putative 1603741 1605384
NMB1549 hypothetical protein 1606176 1606325
NMB1550 conserved hypothetical protein 1606332 1606613
NMB1551 conserved hypothetical protein 1606617 1607717
NMB1552 pilin gene inverting protein PivNM-1A 1608019 1608972
NMB1553 transposase, truncation 1612022 1611708
NMB1554 CTP synthase 1613884 1612253
NMB1555 long-chain-fatty-acid--CoA ligase 1615666 1613999
NMB1556 tRNA (5-methylaminomethyl-2-thiouridylate) -methyltransferase
1616840 1615740
NMB1557 conserved hypothetical protein 1617439 1616969
NMB1558 diacylglycerol kinase 1618115 1617735
NMB1559 glutathione synthetase 1619386 1618430
NMB1560 glutaminyl-tRNA synthetase 1621164 1619479
NMB1561 transcriptional regulator, DeoR family 1622049 1621279
NMB1562 conserved hypothetical protein 1622994 1622095
NMB1563 transcriptional regulator, GntR family 1623859 1623146
NMB1564 conserved hypothetical protein 1624850 1624431
NMB1565 hypothetical protein 1625639 1624971
NMB1566 phosphoribosylglycinamide formyltransferase 1626281 1625658
NMB1567 macrophage infectivity potentiator 1627206 1626391
NMB1568 DNA polymerase holoenzyme chi subunit, putative 1627905 1627468
NMB1569 aminopeptidase A/I, FRAMESHIFT 1629499 1627971
NMB1570 conserved hypothetical protein 1629544 1630656
NMB1571 conserved hypothetical protein 1630656 1631723
NMB1572 aconitate hydratase 2 1631936 1634518
NMB1573 ornithine carbamoyltransferase, catabolic 1634663 1635655
NMB1574 ketol-acid reductoisomerase 1636895 1635885
NMB1575 conserved hypothetical protein 1637268 1636978
NMB1576 acetolactate synthase III, small subunit 1637826 1637338
NMB1577 acetolactate synthase III, large subunit 1639564 1637840
NMB1578 conserved hypothetical protein 1640685 1641335
NMB1579 ATP phosphoribosyltransferase 1641417 1642067
NMB1580 hypothetical protein 1642174 1643070
NMB1581 histidinol dehydrogenase 1643070 1644356
NMB1582 histidinol-phosphate aminotransferase 1644405 1645499
NMB1583 imidazoleglycerol-phosphate dehydratase 1645499 1646413
NMB1584 3-hydroxyacid dehydrogenase 1646511 1647377
NMB1585 transcriptional regulator, MarR family 1647658 1648086
NMB1586 hypothetical protein 1648100 1648963
NMB1587 protease, putative 1650120 1649020
NMB1588 CDP-diacylglycerol--glycerol-3-phosphate 3-phosphatidyltransferase
1651479 1650919
NMB1589 hypothetical protein 1652036 1651797
NMB1590 conserved hypothetical protein 1652675 1652343
NMB1591 transcriptional regulator MtrA 1652804 1653706
NMB1592 hypothetical protein 1653729 1654313
NMB1593 conserved hypothetical protein 1654445 1655305
NMB1594 spermidine/putrescine ABC transporter, periplasmic
spermidine/putrescine-binding protein 1656479 1655352
NMB1595 alanyl-tRNA synthetase 1656684 1659305
NMB1596 hypothetical protein 1659348 1659551
NMB1597 hypothetical protein 1659569 1659997
NMB1598 hypothetical protein 1660094 1660282
NMB1599 hypothetical protein 1660300 1660584
NMB1600 hypothetical protein 1660624 1660878
NMB1601 IS1106 transposase 1661075 1662079
NMB1602 transposase, putative 1663112 1661997
NMB1603 tellurite resistance protein, putative 1663289 1664230
NMB1604 phosphoglycerate mutase 1664989 1664309
NMB1605 topoisomerase IV subunit A 1665137 1667437
NMB1606 sensor histidine kinase 1667460 1669033
NMB1607 sigma-54 dependent response regulator 1669029 1669493
NMB1608 conserved hypothetical protein 1669600 1670349

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NMB1609 trans-sulfuration enzyme family protein 1672860 1671694
NMB1610 hypothetical protein 1673766 1673008
NMB1611 hypothetical protein 1673866 1674114
NMB1612 amino acid ABC transporter, periplasmic amino acid-binding protein
1674169 1674972
NMB1613 fumarate hydratase, class I 1675282 1676802
NMB1614 Trk system potassium uptake protein TrkA 1676903 1678312
NMB1615 hypothetical protein 1678758 1679018
NMB1616 phosphomethylpyrimidine kinase 1679755 1680558
NMB1617 tellurite resistance protein, putative 1681480 1680614
NMB1618 ribonuclease HI 1681594 1682028
NMB1619 conserved hypothetical protein 1682889 1683290
NMB1620 conserved hypothetical protein 1683333 1684514
NMB1621 glutathione peroxidase 1685113 1684583
NMB1622 nitric oxide reductase 1687547 1685295
NMB1623 major anaerobically induced outer membrane protein 1687918 1689087
NMB1624 conserved hypothetical protein 1689215 1689967
NMB1625 pilin gene inverting protein PivNM-1B 1691651 1690698
NMB1626 conserved hypothetical protein 1693053 1691953
NMB1627 conserved hypothetical protein 1693338 1693057
NMB1628 tspB protein, putative 1695347 1693797
NMB1629 Hypothetical protein 1695690 1695328
NMB1630 hypothetical protein 1696057 1695758
NMB1631 hypothetical protein 1696449 1696088
NMB1632 hypothetical protein 1696752 1696555
NMB1633 hypothetical protein 1697067 1696759
NMB1634 conserved hypothetical protein 1698296 1697091
NMB1635 hypothetical protein 1698662 1698444
NMB1636 opacity protein FRAMESHIFT 1700231 1701047
NMB1637 conserved hypothetical protein 1701808 1701254
NMB1638 YhbX/YhjW/YijP/YjdB family protein 1703518 1701887
NMB1639 hypothetical protein 1703921 1703595
NMB1640 phosphoserine aminotransferase 1705027 1703924
NMB1641 conserved hypothetical protein 1705374 1705820
NMB1642 N utilization substance protein A 1705851 1707350
NMB1643 translation initiation factor IF-2 1707365 1710250
NMB1644 hypothetical protein 1711755 1710418
NMB1645 hypothetical protein 1713169 1711832
NMB1646 hemolysin, putative 1713312 1713935
NMB1647 amino acid symporter, putative 1715420 1714005
NMB1648 conserved hypothetical protein 1715747 1716472
NMB1649 disulfide bond formation protein B 1717022 1716537
NMB1650 leucine-responsive regulatory protein 1718177 1717716
NMB1651 alanine racemase 1718502 1719557
NMB1652 conserved hypothetical protein 1720979 1719627
NMB1653 conserved hypothetical protein 1721266 1720997
NMB1654 conserved hypothetical protein 1722129 1721395
NMB1655 adenine specific methylase, putative 1723321 1722413
NMB1656 hypothetical protein 1723454 1724044
NMB1657 comE operon protein 1-related protein 1725327 1724713
NMB1658 DNA/pantothenate metabolism flavoprotein 1731065 1732246
NMB1659 guanosine-3',5'-bis(diphosphate) 3'-pyrophosphohydrolase 1734472
1732319
NMB1660 DNA-directed RNA polymerase, omega subunit 1734770 1734567
NMB1661 guanylate kinase 1735446 1734832
NMB1662 adenine phosphoribosyltransferase 1735607 1736170
NMB1663 conserved hypothetical protein 1737007 1736222
NMB1664 protease, putative 1737332 1738684
NMB1665 conserved hypothetical protein 1739253 1738870
NMB1666 hypothetical protein 1739498 1739253
NMB1667 hypothetical protein 1740061 1739858
NMB1668 hemoglobin receptor 1742596 1740224
NMB1669 iron-starvation protein PigA 1743420 1742794
NMB1670 PqiA family protein 1743706 1745214

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NMB1671 pqiB protein 1745210 1746868
NMB1672 conserved hypothetical protein 1746871 1747386
NMB1673 DNA-3-methyladenine glycosylase I, putative 1747393 1747941
NMB1674 GDSL lipase family protein 1747934 1748572
NMB1675 hypothetical protein 1748797 1749102
NMB1676 glycine dehydrogenase (decarboxylating) 1749136 1751984
NMB1677 cytochrome c5 1753288 1752452
NMB1678 aromatic-amino-acid aminotransferase 1754906 1753716
NMB1679 tRNA (uracil-5-)-methyltransferase 1756015 1754930
NMB1680 chorismate synthase 1756162 1757259
NMB1681 hypothetical protein 1757354 1757776
NMB1682 topoisomerase IV subunit B 1759838 1757856
NMB1683 MutT/nudix family protein 1760429 1759908
NMB1684 seryl-tRNA synthetase 1760595 1761887
NMB1685 D-lactate dehydrogenase 1762966 1761971
NMB1686 peptide chain release factor 1 1764167 1763094
NMB1687 conserved hypothetical protein 1765042 1764275
NMB1688 L-asparaginase I 1766051 1765053
NMB1689 dedA protein, putative 1767007 1766327
NMB1690 phosphoglucomutase/phosphomannomutase family protein 1768532
1767201
NMB1691 dihydropteroate synthase 1769519 1768665
NMB1692 chorismate mutase-related protein 1770552 1769662
NMB1693 hypothetical protein 1770643 1772754
NMB1694 conserved hypothetical protein 1774305 1772824
NMB1695 hypothetical protein 1774424 1775401
NMB1696 acyl carrier protein 1775800 1775558
NMB1697 acyl carrier protein, putative 1776072 1775815
NMB1698 acyltransferase, putative 1776827 1776072
NMB1699 hypothetical protein 1777185 1776823
NMB1700 hypothetical protein 1777345 1777707
NMB1701 hypothetical protein 1777763 1778260
NMB1702 3-oxoacyl-(acyl-carrier-protein) reductase 1778291 1779016
NMB1703 3-oxoacyl-(acyl-carrier-protein) synthase II 1779013 1780260
NMB1704 beta-1,4-glucosyltransferase 1780467 1781222
NMB1705 alpha-1,2-N-acetylglucosamine transferase 1781226 1782287
NMB1706 hypothetical protein 1782329 1782496
NMB1707 sodium- and chloride-dependent transporter 1782677 1784011
NMB1708 NosX-related protein 1784846 1784189
NMB1709 thymidylate synthase 1785648 1784857
NMB1710 glutamate dehydrogenase, NADP-specific 1786032 1787363
NMB1711 transcriptional regulator, GntR family 1788280 1787504
NMB1712 L-lactate permease-related protein 1788711 1789007
NMB1713 transposase, IS30 family 1790361 1789399
NMB1714 multidrug efflux pump channel protein 1791874 1790474
NMB1715 multiple transferable resistance system protein MtrD 1795132
1791932
NMB1716 membrane fusion protein 1796382 1795147
NMB1717 transcriptional regulator MtrR 1796785 1797414
NMB1718 hypothetical protein 1797953 1797699
NMB1719 efflux pump component MtrF 1798240 1799805
NMB1720 exodeoxyribonuclease V 125 kD polypeptide 1803085 1799879
NMB1721 conserved hypothetical protein 1804596 1803190
NMB1722 cytochrome C555 FRAMESHIFT 1804923 1804801
NMB1723 cytochrome c oxidase, subunit III 1806129 1805035
NMB1724 cytochrome c oxidase, subunit II 1806939 1806331
NMB1725 cytochrome c oxidase, subunit I 1808411 1806969
NMB1726 conserved hypothetical protein 1808726 1810471
NMB1727 conserved hypothetical protein 1810539 1810964
NMB1728 biopolymer transport protein ExbD 1812088 1811657
NMB1729 biopolymer transport protein ExbB 1812753 1812094
NMB1730 TonB protein 1813661 1812822
NMB1731 conserved hypothetical protein 1813916 1814551
NMB1732 transporter, putative 1815806 1815009

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NMB1733 hypothetical protein 1816445 1815945
NMB1734 glutaredoxin 1817423 1816785
NMB1735 GTP pyrophosphokinase 1817566 1819776
NMB1736 transposase, putative FRAMESHIFT 1820048 1820856
NMB1737 secretion protein, putative 1822426 1821026
NMB1738 secretion protein, putative 1823922 1822498
NMB1739 hypothetical protein 1824158 1824508
NMB1740 hypothetical protein 1824635 1825042
NMB1741 conserved hypothetical protein FRAMESHIFT 1825116 1826455
NMB1742 hypothetical protein 1826503 1826790
NMB1743 hypothetical protein 1826798 1826992
NMB1744 hypothetical protein 1827003 1827284
NMB1745 hypothetical protein 1827294 1827569
NMB1746 hypothetical protein 1827700 1827987
NMB1747 tspB protein, putative 1828031 1829533
NMB1748 conserved hypothetical protein 1829537 1829824
NMB1749 conserved hypothetical protein 1829837 1830919
NMB1750 pilin gene inverting protein PivNM-2 1831548 1832495
NMB1751 transposase, degenerate 1833264 1832887
NMB1752 conserved hypothetical protein FRAMESHIFT 1833772 1833299
NMB1753 VapD-related protein 1834647 1835081
NMB1754 cryptic plasmid protein A-related protein 1835182 1835084
NMB1755 hypothetical protein 1835328 1835669
NMB1756 hypothetical protein 1835980 1836171
NMB1757 hypothetical protein 1836529 1836756
NMB1758 hypothetical protein 1837008 1837217
NMB1759 conserved hypothetical protein 1837403 1838764
NMB1760 conserved hypothetical protein 1839128 1839631
NMB1761 conserved hypothetical protein 1839797 1841047
NMB1762 hemolysin activation protein HecB, putative 1843162 1841378
NMB1763 toxin-activating protein, putative 1843675 1843220
NMB1764 hypothetical protein 1844155 1843844
NMB1765 hypothetical protein 1844466 1844170
NMB1766 hypothetical protein 1845460 1844450
NMB1767 hypothetical protein 1845945 1845532
NMB1768 hemagglutinin/hemolysin-related protein 1853493 1845952
NMB1769 IS1016 family transposase, putative truncation 1853631 1853822
NMB1770 transposase, IS30 family 1854072 1855034
NMB1771 hypothetical protein 1855539 1855108
NMB1772 hypothetical protein 1857374 1855539
NMB1773 hypothetical protein 1857783 1857412
NMB1774 hypothetical protein 1858438 1858064
NMB1775 hypothetical protein 1860252 1858450
NMB1776 hypothetical protein 1860353 1860252
NMB1777 hypothetical protein 1861364 1861122
NMB1778 hypothetical protein 1861489 1861388
NMB1779 hemagglutinin/hemolysin-related protein 1867499 1861515
NMB1780 hemolysin activation protein HecB, putative 1869350 1867611
NMB1781 hypothetical protein 1869919 1869752
NMB1782 hypothetical protein 1870236 1869937
NMB1783 secretion protein, putative FRAMESHIFT 1871826 1870605
NMB1784 hypothetical protein 1872240 1871890
NMB1785 hypothetical protein 1872472 1872236
NMB1786 hypothetical protein 1873623 1872472
NMB1787 N-acetyl-gamma-glutamyl-phosphate reductase 1874156 1875196
NMB1788 ATP-dependent DNA helicase RecG 1878304 1876265
NMB1789 protein-export protein SecB 1878833 1878393
NMB1790 glutaredoxin 3 1879111 1878857
NMB1791 cytoplasmic axial filament protein FRAMESHIFT 1879236 1880813
NMB1792 sensor histidine kinase 1881795 1880854
NMB1793 response regulator, putative FRAMESHIFT 1882272 1881854
NMB1794 citrate transporter 1883808 1882498
NMB1795 hypothetical protein 1884071 1883916
NMB1796 conserved hypothetical protein 1884950 1884381

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NMB1797 penicillin-binding protein 3 1885109 1886515
NMB1798 IS1016 family transposase, putative FRAMESHIFT 1887236 1886597
NMB1799 S-adenosylmethionine synthetase 1888654 1887488
NMB1800 hypothetical protein 1888703 1888903
NMB1801 HtrB/MsbB family protein 1889000 1889893
NMB1802 O-sialoglycoprotein endopeptidase 1891004 1889943
NMB1803 cytochrome c-type biogenesis protein, putative 1892308 1891124
NMB1804 cytochrome c-type biogenesis protein, putative 1894316 1892304
NMB1805 cytochrome c4 1895153 1894533
NMB1806 conserved hypothetical protein 1895353 1895985
NMB1807 penicillin-binding protein 1 1898505 1896112
NMB1808 pilM protein 1898657 1899769
NMB1809 pilN protein FRAMESHIFT 1899775 1900371
NMB1810 pilO protein 1900375 1901019
NMB1811 pilP protein 1901040 1901582
NMB1812 pilQ protein FRAMESHIFT 1901604 1903908
NMB1813 shikimate kinase 1904813 1905322
NMB1814 3-dehydroquinate synthase 1905405 1906481
NMB1815 conserved hypothetical protein 1907451 1908290
NMB1816 conserved hypothetical protein 1908323 1908784
NMB1817 riboflavin-specific deaminase 1908819 1909925
NMB1818 lipopolysaccharide biosynthesis protein, putative 1910123 1911541
NMB1819 hypothetical protein 1911541 1911693
NMB1820 pilin glycosylation protein PglB 1911712 1912950
NMB1821 pilin glycosylation protein PglC 1913086 1914258
NMB1822 pilin glycosylation protein PglD 1914309 1916216
NMB1823 valine--pyruvate aminotransferase 1916275 1917564
NMB1824 conserved hypothetical protein 1918455 1917622
NMB1825 hypothetical protein 1919103 1918903
NMB1826 conserved hypothetical protein 1919452 1919084
NMB1827 DNA polymerase III, alpha subunit 1919852 1923283
NMB1828 conserved hypothetical protein 1924652 1923723
NMB1829 TonB-dependent receptor 1926848 1924725
NMB1830 phosphoglycolate phosphatase, putative 1926996 1927652
NMB1831 lytB protein 1928711 1927746
NMB1832 lipoprotein signal peptidase 1929267 1928743
NMB1833 isoleucyl-tRNA synthetase 1933332 1930546
NMB1834 riboflavin kinase/FMN adenylyltransferase 1934394 1933477
NMB1835 tyrosyl-tRNA synthetase 1936217 1934925
NMB1836 lipopolysaccharide biosynthesis protein WbpC, putative 1938151
1936283
NMB1837 hypothetical protein 1938466 1938215
NMB1838 GTP-binding protein, putative 1939615 1938527
NMB1839 formate--tetrahydrofolate ligase 1941406 1939733
NMB1840 conserved hypothetical protein 1941581 1942009
NMB1841 mannose-1-phosphate guanylyltransferase-related protein 1942741
1942049
NMB1842 4-hydroxyphenylacetate 3-hydroxylase, small subunit, putative
1943257 1942760
NMB1843 transcriptional regulator, MarR family 1943812 1943375
NMB1844 hypothetical protein 1943938 1943819
NMB1845 thioredoxin 1944662 1944156
NMB1846 Mrp/NBP35 family protein 1945032 1946108
NMB1847 pilC1 protein FRAMESHIFT 1947287 1950374
NMB1848 hypothetical protein 1952279 1951938
NMB1849 carbamoyl-phosphate synthase, small subunit 1952589 1953719
NMB1850 hypothetical protein 1954091 1954363
NMB1851 hypothetical protein 1954440 1954697
NMB1852 conserved hypothetical protein 1954697 1955083
NMB1853 hypothetical protein 1955422 1955691
NMB1854 hypothetical protein 1955768 1956406
NMB1855 carbamoyl-phosphate synthase, large subunit 1956438 1959650
NMB1856 transcriptional regulator, LysR family 1960777 1959881
NMB1857 modulator of drug activity B 1961016 1961591

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NMB1858 hypothetical protein 1961977 1961594
NMB1859 S-adenosylmethionine:tRNA ribosyltransferase-isomerase 1963108
1962071
NMB1860 acetyl-CoA carboxylase, biotin carboxyl carrier protein 1963464
1963916
NMB1861 acetyl-CoA carboxylase, biotin carboxylase 1964031 1965389
NMB1862 ribosomal protein L11 methyltransferase 1965653 1966537
NMB1863 oligoribonuclease 1966558 1967118
NMB1864 glutamate-1-semialdehyde 2,1-aminomutase 1968808 1967528
NMB1865 hypothetical protein 1968821 1969036
NMB1866 conserved hypothetical protein 1969593 1970918
NMB1867 1-deoxyxylulose-5-phosphate synthase 1972919 1971009
NMB1868 integrase/recombinase XerC 1973909 1973007
NMB1869 fructose-bisphosphate aldolase 1974093 1975154
NMB1870 hypothetical protein 1975177 1976136
NMB1871 conserved hypothetical protein 1976286 1976960
NMB1872 ribosomal-protein-alanine acetyltransferase, putative 1976960
1977397
NMB1873 DNA polymerase, bacteriophage-type, putative 1977394 1978128
NMB1874 orotate phosphoribosyltransferase 1978193 1978831
NMB1875 hypothetical protein 1978908 1979339
NMB1876 N-acetylglutamate synthase 1979339 1980646
NMB1877 prolyl oligopeptidase family protein 1980850 1982862
NMB1878 transcriptional regulator, AraC family 1983567 1982983
NMB1879 hypothetical protein 1983936 1983628
NMB1880 ABC transporter, periplasmic solute-binding protein, putative
1984172 1985134
NMB1881 conserved hypothetical protein 1985694 1986014
NMB1882 TonB-dependent receptor 1986131 1988305
NMB1883 hypothetical protein 1988727 1988440
NMB1884 conserved hypothetical protein 1989047 1988727
NMB1885 protein-L-isoaspartate O-methyltransferase 1989783 1989130
NMB1886 conserved hypothetical protein 1990389 1989889
NMB1887 triosephosphate isomerase 1990568 1991338
NMB1888 protein-export membrane protein SecE 1991348 1991695
NMB1889 hypothetical protein 1992486 1992575
NMB1890 conserved hypothetical protein 1992709 1993074
NMB1891 helix-turn-helix family protein 1993074 1993382
NMB1892 hypothetical protein 1993495 1993704
NMB1893 conserved hypothetical protein FRAMESHIFT 1994615 1993771
NMB1894 leucyl-tRNA synthetase, truncation 1994851 1994723
NMB1895 DNA adenine methylase, truncation 1994987 1994847
NMB1896 type II restriction enzyme DpnI 1995774 1994974
NMB1897 leucyl-tRNA synthetase 1998538 1995911
NMB1898 lipoprotein 1998808 1999320
NMB1899 hypothetical protein 1999330 1999770
NMB1900 polyphosphate kinase 1999849 2001996
NMB1901 IS1016C2 transposase, degenerate 2002232 2002770
NMB1902 DNA polymerase III, beta subunit 2004113 2003013
NMB1903 chromosomal replication initiator protein DnaA 2005904 2004351
NMB1904 ribosomal protein L34 2006196 2006327
NMB1905 ribonuclease P protein component 2006333 2006695
NMB1906 conserved hypothetical protein 2006763 2006981
NMB1907 60 kd inner-membrane protein 2007156 2008790
NMB1908 conserved hypothetical protein 2009599 2008877
NMB1909 Maf/YceF/YhdE family protein 2010236 2009649
NMB1910 conserved hypothetical protein 2010384 2010884
NMB1911 50S ribosomal protein L32 2010921 2011097
NMB1912 conserved hypothetical protein 2011275 2011799
NMB1913 fatty acid/phospholipid synthesis protein 2011891 2012943
NMB1914 hypothetical protein 2013082 2013330
NMB1915 hypothetical protein 2013360 2013746
NMB1916 3-oxoacyl-(acyl-carrier-protein) synthase III 2013931 2014890
NMB1917 conserved hypothetical protein 2014940 2015344

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NMB1918 malonyl CoA-acyl carrier protein transacylase 2015441 2016364
NMB1919 ABC transporter, ATP-binding protein 2016505 2018367
NMB1920 GMP synthase 2018470 2020032
NMB1921 3-oxoacyl-(acyl-carrier-protein) reductase 2020097 2020840
NMB1922 IS1106 transposase, degenerate 2021273 2021118
NMB1923 conserved hypothetical protein 2021377 2021757
NMB1924 inositol monophosphatase family protein 2022673 2021981
NMB1925 conserved hypothetical protein 2022876 2023598
NMB1926 lacto-N-neotetraose biosynthesis glycosyl transferase LgtE 2025680
2024841
NMB1927 lacto-N-neotetraose biosynthesis glycosyl transferase-related
protein 2025817 2025725
NMB1928 lacto-N-neotetraose biosynthesis glycosyl transferase LgtB 2026656
2025832
NMB1929 lacto-N-neotetraose biosynthesis glycosyl transferase LgtA 2027747
2026701
NMB1930 glycyl-tRNA synthetase, beta chain 2029827 2027767
NMB1931 hypothetical protein 2030256 2029912
NMB1932 glycyl-tRNA synthetase, alpha chain 2031238 2030336
NMB1933 ATP synthase F1, epsilon subunit 2032065 2031646
NMB1934 ATP synthase F1, beta subunit 2033473 2032079
NMB1935 ATP synthase F1, gamma subunit 2034386 2033514
NMB1936 ATP synthase F1, alpha subunit 2035958 2034414
NMB1937 ATP synthase F1, delta subunit 2036502 2035972
NMB1938 ATP synthase F0, B subunit 2036977 2036510
NMB1939 ATP synthase F0, C subunit 2037284 2037051
NMB1940 ATP synthase F0, A subunit 2038207 2037344
NMB1941 hypothetical protein 2038550 2038200
NMB1942 hypothetical protein 2038997 2038707
NMB1943 hypothetical protein 2039340 2039170
NMB1944 ParB family protein 2040252 2039395
NMB1945 3-octaprenyl-4-hydroxybenzoate carboxy-lyase 2040407 2040976
NMB1946 outer membrane lipoprotein 2041904 2041044
NMB1947 ABC transporter, permease protein 2042749 2042066
NMB1948 ABC transporter, ATP-binding protein 2043488 2042754
NMB1949 soluble lytic murein transglycosylase, putative 2044018 2045865
NMB1950 30S ribosomal protein S21 2046157 2046366
NMB1951 conserved hypothetical protein 2046405 2046944
NMB1952 stringent starvation protein B 2047538 2047149
NMB1953 stringent starvation protein A 2048215 2047613
NMB1954 hypothetical protein 2050146 2048488
NMB1955 cadmium resistance protein 2050933 2050310
NMB1956 50S ribosomal protein L31 2051451 2051239
NMB1957 acetyltransferase-related protein FRAMESHIFT 2051688 2052197
NMB1958 thioredoxin, putative 2052770 2052273
NMB1959 conserved hypothetical protein 2053150 2052770
NMB1960 hypothetical protein 2053632 2053153
NMB1961 VacJ-related protein 2054464 2053640
NMB1962 hypothetical protein 2054739 2054464
NMB1963 conserved hypothetical protein 2055380 2054793
NMB1964 conserved hypothetical protein 2055911 2055420
NMB1965 conserved hypothetical protein 2056738 2055965
NMB1966 ABC transporter, ATP-binding protein 2057586 2056789
NMB1967 transcriptional regulator, AraC family 2057759 2058673
NMB1968 aldehyde dehydrogenase A 2058936 2060375
NMB1969 serotype-1-specific antigen, putative 2061412 2064657
NMB1970 para-aminobenzoate synthetase component I/4-amino-4-
deoxychorismate lyase, putative 2065692 2067470
NMB1971 conserved hypothetical protein 2069049 2067535
NMB1972 chaperonin, 60 kDa 2071379 2069748
NMB1973 chaperonin, 10 kDa 2071762 2071475
NMB1974 IS1016C2 transposase, degenerate 2071990 2072639
NMB1975 sodium- and chloride-dependent transporter 2072855 2074387
NMB1976 diaminopimelate decarboxylase 2075759 2074518

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NMB1977 hypothetical protein 2075940 2075773
NMB1978 cyaY protein 2076011 2076331
NMB1979 conserved hypothetical protein 2076361 2077374
NMB1980 conserved hypothetical protein 2077403 2077819
NMB1981 conserved hypothetical protein 2077844 2078347
NMB1982 DNA polymerase I 2078496 2081309
NMB1983 hypothetical protein 2082658 2083326
NMB1984 IS1106 transposase FRAMESHIFT 2083391 2084499
NMB1985 adhesion and penetration protein 2089191 2084821
NMB1986 hypothetical protein 2089756 2089328
NMB1987 thiophene and furan oxidation protein ThdF 2090041 2091384
NMB1988 iron-regulated outer membrane protein FrpB 2092611 2094752
NMB1989 iron(III) ABC transporter, periplasmic binding protein 2095472
2096434
NMB1990 iron(III) ABC transporter, permease protein 2096601 2097566
NMB1991 iron(III) ABC transporter, permease protein 2097559 2098530
NMB1992 hypothetical protein 2098577 2099200
NMB1993 iron(III) ABC transporter, ATP-binding protein 2099286 2100041
NMB1994 adhesin/invasin, putative 2100342 2101433
NMB1995 nitrogen regulatory protein P-II, FRAMESHIFT 2101839 2101423
NMB1996 phosphoribosylformylglycinamidine synthase 2101990 2105949
NMB1997 hydroxyacylglutathione hydrolase 2106047 2106802
NMB1998 serine-type peptidase 2107119 2111411
NMB1999 magnesium transporter 2111646 2113097
NMB2000 conserved hypothetical protein 2114094 2113189
NMB2001 conserved hypothetical protein 2114339 2115091
NMB2002 hypothetical protein 2115113 2115328
NMB2003 conserved hypothetical protein 2115476 2115820
NMB2004 conserved hypothetical protein 2115820 2116509
NMB2005 glutamate N-acetyltransferase/amino-acid acetyltransferase 2116579
2117796
NMB2006 chloride channel protein-related protein 2117859 2119265
NMB2007 ATP-dependent RNA helicase HrpA, truncation 2119458 2120846
NMB2008 ABC transporter, ATP-binding protein-related protein 2120993
2122633
NMB2009 ATP-dependent RNA helicase HrpA, degenerate 2122680 2122859
NMB2010 YhbX/YhjW/YijP/YjdB family protein 2123074 2124648
NMB2011 ATP-dependent RNA helicase HrpA, truncation 2124717 2128133
NMB2012 transcriptional regulator, HTH 3 family 2129260 2128172
NMB2013 hypothetical protein 2129920 2129279
NMB2014 hypothetical protein 2130249 2130004
NMB2015 hypothetical protein 2130614 2130880
NMB2016 type IV pilin-related protein 2131493 2131047
NMB2017 ComEA-related protein 2132027 2131584
NMB2018 conserved hypothetical protein 2138411 2137752
NMB2019 lipopolysaccharide core biosynthesis protein KdtB 2138949 2138440
NMB2020 conserved hypothetical protein 2139756 2139076
NMB2021 conserved hypothetical protein 2140179 2139916
NMB2022 conserved hypothetical protein 2140722 2140255
NMB2023 conserved hypothetical protein 2141162 2140779
NMB2024 conserved hypothetical protein 2141826 2141224
NMB2025 conserved hypothetical protein 2142422 2141826
NMB2026 ABC transporter, permease protein 2144046 2142454
NMB2027 gluconate permease 2144385 2145767
NMB2028 thermoresistant gluconokinase 2145790 2146305
NMB2029 homoserine kinase FRAMESHIFT 2147564 2146650
NMB2030 3-demethylubiquinone-9 3-methyltransferase 2148329 2147604
NMB2031 tryptophan transporter 2148481 2149719
NMB2032 lipopolysaccharide glycosyl transferase, FRAMESHIFT 2149872
2150922
NMB2033 histidinol-phosphatase, putative 2151173 2151733
NMB2034 1-acyl-sn-glycerol-3-phosphate acyltransferase, putative 2151765
2152505
NMB2035 conserved hypothetical protein 2152505 2153194

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NMB2036 tRNA pseudouridine synthase A 2154495 2155390
NMB2037 hypothetical protein 2155415 2155651
NMB2038 PemK-related protein 2155642 2155962
NMB2039 major outer membrane protein PIB 2157487 2158479
NMB2040 thiamine biosynthesis protein ThiC 2161479 2159581
NMB2041 thiamin pyrophosphokinase-related protein 2162093 2162965
NMB2042 spermidine/putrescine ABC transporter, ATP-binding protein 2162977
2163912
NMB2043 IS1106 transposase, putative POINT MUTATION 2165702 2164734
NMB2044 phosphoenolpyruvate-protein phosphotransferase 2168278 2166506
NMB2045 phosphocarrier protein HPr 2168547 2168281
NMB2046 PTS system, IIAB component 2169074 2168619
NMB2047 hypoxanthine-guanine phosphoribosyltransferase, putative 2169697
2169137
NMB2048 DNA ligase 2170590 2169769
NMB2049 glyoxalase II family protein 2170682 2171311
NMB2050 conserved hypothetical protein 2173305 2171524
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NMB2052 ubiquinol--cytochrome c reductase, cytochrome b 2175793 2174447
NMB2053 ubiquinol--cytochrome c reductase, iron-sulfur subunit 2176393
2175815
NMB2054 conserved hypothetical protein 2177265 2176519
NMB2055 transcriptional regulator, LysR family 2177396 2178322
NMB2056 30S ribosomal protein S9 2178972 2178583
NMB2057 50S ribosomal protein L13 2179413 2178985
NMB2058 conserved hypothetical protein 2180081 2179779
NMB2059 hypothetical protein 2180421 2180095
NMB2060 glycerol-3-phosphate dehydrogenase (NAD+) 2181465 2180479
NMB2061 phosphoenolpyruvate carboxylase 2184290 2181591
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NMB2063 slyX protein, putative 2186018 2185797
NMB2064 conserved hypothetical protein 2187407 2186022
NMB2065 hemK protein FRAMESHIFT 2188764 2187496
NMB2066 tldD protein 2190271 2188832
NMB2067 conserved hypothetical protein 2190661 2191881
NMB2068 D-amino acid oxidase flavoprotein, putative 2191881 2192978
NMB2069 thiamin-phosphate pyrophosphorylase 2193003 2193617
NMB2070 hypothetical protein 2194042 2194233
NMB2071 thiG protein 2194450 2195235
NMB2072 hypothetical protein 2195352 2195492
NMB2073 hypothetical protein 2195580 2195780
NMB2074 hypothetical protein 2196867 2196004
NMB2075 BirA protein/Bvg accessory factor 2198657 2196882
NMB2076 aut protein 2199160 2198657
NMB2077 methylenetetrahydrofolate dehydrogenase/methenyltetrahydrofolate
cyclohydrolase FRAMESHIFT 2199800 2200650
NMB2078 conserved hypothetical protein 2201296 2200718
NMB2079 aspartate-semialdehyde dehydrogenase 2201472 2202584
NMB2080 hypothetical protein 2203345 2202818
NMB2081 hypothetical protein 2203700 2203359
NMB2082 exodeoxyribonuclease 2204466 2203690
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NMB2085 hypothetical protein 2207707 2206661
NMB2086 GTP-binding protein 2208944 2207793
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NMB2088 conserved hypothetical protein 2210766 2209894
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NMB2090 phosphoheptose isomerase 2211164 2211754
NMB2091 hemolysin, putative 2211821 2212426
NMB2092 hypothetical protein 2212437 2213066
NMB2093 methionine aminopeptidase 2213109 2213885
NMB2094 hypothetical protein 2214043 2214339
NMB2095 adhesin complex protein, putative 2214580 2214951

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NMB2096 malate:quinone oxidoreductase 2216608 2215145
NMB2097 hypothetical protein 2216749 2216663
NMB2098 conserved hypothetical protein 2217735 2217148
NMB2099 conserved hypothetical protein 2218377 2217799
NMB2100 hypothetical protein 2218455 2218685
NMB2101 30S ribosomal protein S2 2218861 2219586
NMB2102 elongation factor TS (EF-TS) 2219718 2220569
NMB2103 uridylate kinase 2220789 2221505
NMB2104 mafA protein FRAMESHIFT 2221692 2222652
NMB2105 mafB protein 2222695 2224143
NMB2106 hypothetical protein 2224143 2224496
NMB2107 MafB-related protein 2224527 2225288
NMB2108 hypothetical protein 2225301 2225504
NMB2109 hypothetical protein 2225639 2225887
NMB2110 hypothetical protein 2225887 2226255
NMB2111 MafB-related protein 2226268 2227110
NMB2112 hypothetical protein 2227306 2227572
NMB2113 hypothetical protein 2227598 2227897
NMB2114 MafB-related protein 2227948 2228583
NMB2115 hypothetical protein 2228589 2228930
NMB2116 hypothetical protein 2228971 2229312
NMB2117 MafB-related protein, degenerate 2229645 2230340
NMB2118 hypothetical protein 2230340 2230654
NMB2119 MafB-related protein 2230709 2231464
NMB2120 hypothetical protein 2231471 2231869
NMB2121 hypothetical protein 2232031 2232372
NMB2122 MafB-related protein 2232409 2232510
NMB2123 hypothetical protein 2232518 2232871
NMB2124 hypothetical protein 2232922 2233047
NMB2125 hypothetical protein 2233047 2233418
NMB2126 IS1016 family transposase, putative FRAMESHIFT 2234296 2233462
NMB2127 protease, putative 2235364 2234381
NMB2128 CinA-related protein 2236204 2235407
NMB2129 argininosuccinate synthase 2236517 2237857
NMB2130 hypothetical protein 2237908 2238147
NMB2131 hypothetical protein 2238143 2238355
NMB2132 transferrin-binding protein-related protein 2239900 2238437
NMB2133 sodium/dicarboxylate symporter family protein 2241384 2240158
NMB2134 conserved hypothetical protein 2241857 2243761
NMB2135 conserved hypothetical protein 2243771 2247985
NMB2136 peptide transporter 2249471 2250925
NMB2137 hypothetical protein 2251451 2251660
NMB2138 peptide chain release factor 2 2252924 2251824
NMB2139 conserved hypothetical protein 2253920 2253030
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NMB2141 hypothetical protein 2254787 2255092
NMB2142 conserved hypothetical protein 2255187 2256050
NMB2143 conserved hypothetical protein 2256043 2256786
NMB2144 sigma factor, putative 2256811 2257395
NMB2145 hypothetical protein 2257404 2257580
NMB2146 hypothetical protein 2257703 2257810
NMB2147 hypothetical protein 2257842 2258261
NMB2148 transposase, IS30 family 2258738 2259700
NMB2149 hypothetical protein 2260052 2259795
NMB2150 conserved hypothetical protein 2261006 2260440
NMB2151 phosphoribosylamine--glycine ligase 2262344 2261076
NMB2152 hypothetical protein 2262502 2262816
NMB2153 conserved hypothetical protein 2263482 2262874
NMB2154 electron transfer flavoprotein, alpha subunit 2264480 2263548
NMB2155 electron transfer flavoprotein, beta subunit 2265240 2264494
NMB2156 heptosyltransferase I 2266435 2265470
NMB2157 pyrazinamidase/nicotinamidase PncA, putative 2267107 2266475
NMB2158 conserved hypothetical protein 2267221 2267898
NMB2159 glyceraldehyde 3-phosphate dehydrogenase 2269163 2268162

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NMB2160 DNA mismatch repair protein Muts 2269607 2272198
NMB0505 hypothetical protein 533467 533186
NMB1123 hypothetical protein 1135584 1135390
NMB1124 hypothetical protein 1136271 1135627
NMB1125 hypothetical protein 1136639 1136271
NMB1126 hypothetical protein 1137317 1136649
NMB1127 oxidoreductase, short chain dehydrogenase/reductase family 1138201
1137485
NMB1129 hypothetical protein 1139833 1139630
NMB1130 phytoene synthase, putative 1140867 1139998
NMB1133 conserved hypothetical protein / ankyrin-related protein 1144428
1143670
NMB1134 ferredoxin, 2Fe-2S type 1144824 1144486
NMB1135 hypothetical protein 1145242 1145102
NMB1137 conserved hypothetical protein 1146211 1146017
NMB1138 conserved hypothetical protein 1146683 1146285
NMB1141 RNA methyltransferase, TrmH family 1150088 1149480
NMB1142 hypothetical protein 1150375 1150142
NMB1143 hypothetical protein 1150909 1150547
NMB1144 hypothetical protein 1151226 1150924, lipoprotein
NMB1147 hypothetical protein 1154639 1154007, homology to plasmid proteins
Y4SH_RISHN and PXO2_BACAN
NMB1149 hypothetical protein 1155016 1154876
NMB1151 sulfite reductase hemoprotein, beta-component 1159086 1157320
NMB1152 sulfite reductase (NADPH) flavoprotein, alpha component 1160927
1159116
NMB1154 sulfate adenylyltransferase, subunit 2 1163172 1162252
NMB1156 siroheme synthase 1165412 1163964
NMB1157 hypothetical protein 1165696 1165541
NMB1159 conserved hypothetical protein 1167316 1166429, inner membrane
NMB1160 conserved hypothetical protein 1167316 1166429
NMB1166 conserved hypothetical protein 1171633 1170323
NMB1169 chaperone protein HscA 1174933 1173074
NMB1170 hypothetical protein 1175666 1175013
NMB1174 hypothetical protein 1178053 1177373
NMB1177 acetyl-CoA carboxylase, carboxyl transferase alpha subunit 1179887
1178931
NMB1178 mesJ protein FRAMESHIFT 1181265 1179984
NMB1183 UDP-N-acetylmuramate:L-alanyl-gamma-D-glutamyl-meso-
diaminopimelate ligase 1184700 1183327
NMB1184 biotin synthetase 1185959 1184910
NMB1186 hypothetical protein 1186881 1186729
NMB1188 dihydroxy-acid dehydratase 1189180 1187324
NMB1191 sulfate adenylyltransferase, subunit 1 1194246 1192963
NMB1193 phosphoadenosine phosphosulfate reductase 1195986 1195249
NMB1196 nickel-dependent hydrogenase, b-type cytochrome subunit 1198401
1197748

INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 00/05928

A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12Q1/68 C12N15/11 C07K14/22

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12Q C12N C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, CHEM ABS Data, MEDLINE, EMBASE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 98 17805 A (RAYMOND NIGEL ;QUINN FREDERICK D (US); US HEALTH (US); RIBOT EFRAI) 30 April 1998 (1998-04-30) the whole document ---	1-4, 7-14, 18-24
X	EP 0 467 714 A (MERCK & CO INC) 22 January 1992 (1992-01-22) claims; example 3 --- -/--	1-4, 7-14, 18-24

☒ Further documents are listed in the continuation of box C.☒ Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

Date of the actual completion of the international search

10 October 2000

Date of mailing of the international search report

19.10.00

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INTERNATIONAL SEARCH REPORT

International Application No

PCT/US 00/05928

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	FLEISCHMANN R D ET AL: "WHOLE-GENOME RANDOM SEQUENCING AND ASSEMBLY OF HAEMOPHILUS INFLUENZAE RD" SCIENCE,US,AMERICAN ASSOCIATION FOR THE ADVANCEMENT OF SCIENCE,, vol. 269, no. 5223, 28 July 1995 (1995-07-28), pages 496-498,507-51, XP000517090 ISSN: 0036-8075 the whole document	1-4, 7-14, 16-24
T	TETTELIN H ET AL: "Complete genome sequence of Neisseria meningitidis serogroup B strain MC58 'see comments!." SCIENCE, (2000 MAR 10) 287 (5459) 1809-15., XP000914963 page 963	
T	PIZZA M ET AL: "Identification of vaccine candidates against serogroup B meningococcus by whole- genome sequencing 'see comments!." SCIENCE, (2000 MAR 10) 287 (5459) 1816-20., XP000914964 the whole document	
T	PARKHILL J ET AL: "Complete DNA sequence of a serogroup A strain of Neisseria meningitidis Z2491 'see comments!." NATURE, (2000 MAR 30) 404 (6777) 502-6., XP000918875 the whole document	

INTERNATIONAL SEARCH REPORT

national application No.
PCT/US 00/05928**Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)**

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.: 16,17 (partly)
because they relate to subject matter not required to be searched by this Authority, namely:
Rule 39.1(v) PCT – Presentation of information (insofar as related to computer databases)
2. ☒ Claims Nos.: 5,6,15 (completely), 1-4, 7-14, 16-24 (partly)
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
see FURTHER INFORMATION sheet PCT/ISA/210
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Claims Nos.: 5,6,15 (completely), 1-4, 7-14, 16-24 (partly)

1) Claims 5 and 6 (and thus 15 which refers to claim 6 and whose reference to claims 7 and 8 is wrong) lack any essential technical feature which could allow a meaningful search to be carried out. They have thus not been searched. For the same reason claims 18-24 have not been searched insofar as referring to any of claims 5, 6 and 15.

2) Claims 1-4, 7-14, 16-24 have only been searched insofar as related to the full sequence SEQ ID 1 in view of the absence of any indication in the claims as to searchable SEQ IDs corresponding to the "NMB open reading frames". SEQ ID 1 as such is not searchable by means of similarity algorithms since it is too long: the search with respect thereto has thus been carried out based on keywords.

3) A further reason for not searching claims 1-4 insofar as related to "NMB open reading frames" is that claim 1 is unclear (Art. 6 PCT). It relates to a method for searching open reading frames "within one or more...NMB open reading frames", which is however technically meaningless.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/US 00/05928

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
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EP 0467714 A	22-01-1992	AU 8114091 A	23-01-1992
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